ASCORBIC ACID THERAPY IN CERTAIN ZYMOTIC DISEASES: A STUDY OF FIVE HUNDRED AND EIGHT CASES, WITH A CRITICAL REVIEW OF THE LITERATURE.

BY

IAN ALEXIS GORDON MACQUEEN,

M.A., M.B., Ch.B., D.P.H.

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INTRODUCTION.

" Much of the medical research of recent generations, especially in Britain, has lost much, and in some cases all, of its value from failure to confirm the facts upon which it is founded; it has wasted time and effort in proving or disproving theories which were not serious and in explaining phenomena which had not been proved to exist." (1)*

The above generalisation, from a leading article in one of last year's journals, is particularly applicable to a goodly portion of the work that has been done on the relationship between vitamin C and infectious diseases. The view that in fevers there is an increased need for vitamin C has gained wide currency, and has even been stated dogmatically in an editorial in 'The Lancet' (2); yet a mere glance at the steadily accumulating mass of literature on the subject reveals that some workers seek to bring about a state of hypervitaminosis, some wish to remedy a hypovitaminosis which they regard as important in the causation of the illness, some maintain that the hypovitaminosis is simply a result of either increased metabolism or reduced intake, and a few deny the very existance of any hypovitaminosis. Even a casual survey, too, makes it obvious that some of the conclusions of early workers in the field were based on methods which have since been proved faulty, or on arbitrary and quite unjustified standards, and that these invalid conclusions have sometimes been accepted by later researchers as a stable basis for their own work.

In these circumstances a comprehensive investigation of the effects of ascorbic acid therapy in infectious diseases seems very desirable.

Part 2 of this thesis is a humble attempt at such an investigation in a few selected diseases, the method employed consisting of a comparative study of patients treated with vitamin C and controls suffering from the same diseases; and Part 1 is a brief critical review of the relevant literature.

PART /

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* For references see Page 92.

PART 1 - REVIEW OF THE LITERATURE.

It is convenient to divide previous investigations of the relationship between vitamin C and infectious diseases into three groups: first, those concerned with determining the effects of infectious conditions on the bodily stores of the vitamin, i.e.,the evidence for and against the existence of hypovitaminosis C in patients suffering from zymotic diseases; second, anti-bacterial effects of ascorbic acid in vitro, and experiments on laboratory animals; and, third, effects of administration of the vitamin to infectious patients.

The first of these three divisions presupposes the existence both of methods of estimating the vitamin and of standards of normality. Since the employment of an inaccurate method of estimation, or the comparison of correct estimates of the vitamin C levels of infectious patients with false standards of normality, might well lead to the formation of erroneous conclusions regarding the presence or absence of hypovitaminosis C, it seems desirable that the methods of assessing the nutritional status in respect of vitamin C and the various standards of normality should also be considered.

The following critical summary of previous work is therefore divided into five sections:

(1) Vitamin C, - methods of assay and normal standards;
(2) Application of these standards to infections;
(3) Effects of ascorbic acid in vitro, and experiments on laboratory animals;
(4) Effects of administration of vitamin C to infectious patients; and
(5) Other points of interest.

(1) VITAMIN C, - METHODS OF ASSAY, AND NORMAL STANDARDS.

The methods of estimating vitamin C include tests of capillary fragility, quantitative estimation of the urinary excretion, "Saturation" tests, an intradermal test, and quantitative estimations of the concentration in blood or plasma.

(A) Capillary Fragility Tests.

A method of estimating vitamin C nutrition by placing a pressure cuff round the arm for a specified time and then counting the number of petechiae that developed in an area of standard size was originated by Hess (3) in 1914, and modifications were introduced by Gothlin (4), Dalldorf and Russell/

Russell (5), Schultz (6), and Brown and Wasson (7). Some lack of relationship between cuff test results and vitamin C was noted by many workers - e.g. Greene (8), O'Hara and Hauch (9), van Eekelen (10), and Farmer (11); and cases were reported of actual scurvy in which the capillary fragility was within normal limits (12 & 13); but it fell to the man who had originally isolated vitamin C, Szent-Gyorgyi, to discover (14) that the permeability of capillaries was controlled, not by vitamin C, but by a previously unknown substance which the Hungarian scientist named, " Vitamin P." The final coup de grace to the cuff test as a measure of vitamin C nutrition was dealt by Scarborough (15), who proved conclusively that ascorbic acid does not decrease capillary fragility, whereas a Hesperidin preparation (i.e. Szent-Gyorgyi's Vitamin P) does so very dramatically.

Capillary fragility tests have been extensively used, even after the discovery of vitamin P. For instance, in 1942 they were employed by Munro, Lazarus, and Bell (12) in the expectation that they would be " the most sensitive index of the state of nutrition in regard to vitamin C "; and, a little earlier, they were used by Boje (16) in his investigation of the prevalence of hypovitaminosis C in Greenland.

Since capillary fragility is definitely not an index of the state of ascorbic acid nutrition, it follows that conclusions based on cuff tests are invalid.

The first standards for the necessary daily intake of vitamin C were formulated by Gothlin (4) in 1931; he found that 21 - 30 mg. were required to " prevent undue capillary fragility"; and in 1937, on the basis of his previous findings, together with certain data from some animal experiments, he estimated (17) that an average adult required 19 - 27 mg. of ascorbic acid daily. These standards, based on an invalid test, have no significance; yet, as Godber (18) has pointed out in a brilliant thesis (which will be discussed later in some detail), they have been made the basis for some subsequent estimates of human requirements of ascorbic acid.

(B) Quantitative Estimates of Vitamin C in Urine.

A quantitative test for vitamin C, based on the principle that the blue dye, 2:6 dichlorophenol-indophenol, is, in acid solution, reduced from a pink to a colourless liquid by ascorbic acid, was devised by Tillmans (19) as early as 1932, and elaborated by Abbasy et alii (20). The dye test is already sufficiently well known to be included in standard text-books on biochemistry (21), and need not, therefore, be described here.

One or two modifications have been suggested, - e.g. Hess /

Hess and Benjamin (25) used the dye test but preserved the urine with trichloroacetic acid, instead of with glacial acetic; Iancou, Oprisiu and Jula (26) advised some changes in the technique; and Harrison (22) suggested regenerating oxidised ascorbic acid with H_2S prior to titration. None of these modifications, however, has passed into popular use. Again, several alternative tests have been devised: the methods of Evelyn (27) and Bessay (28), both involving the use of a photo-electric colorimeter, are fairly specific but unsuitable for general use; that of Medes (29 & 30), titrating with phospho-18-tungstic acid, was claimed by the authoress to be more accurate for urines rich in ascorbic acid, but is too complicated for general clinical work; and that of Roe and Hall (31), using a 2:4 dinitrophenylhydrazine derivative of dehydroascorbic acid, does not appear to have any great advantage over the original dye test. Prior to 1941, then, the dichlorophenol-indophenol test stood virtually unchallenged.

In 1941, G. T. Meiklejohn and C. P. Stewart devised an entirely new test for ascorbic acid. Their method, based on the fact that cucumber seeds contain an active enzyme that oxidises vitamin C, is fairly simple, involves no elaborate apparatus, and - unlike previous methods - is absolutely specific for ascorbic acid. Hence it is likely to become the method of choice in the future.

However, the enzyme test is still comparatively new. Practically all the work on the excretion of vitamin C has been done with the dichlorophenol-indophenol test, the accuracy of which must now be considered.

The objection that it is seldom convenient to titrate every specimen of urine immediately it is voided, and that in stored urine a portion of the ascorbic acid is lost by oxidation, carries little weight; for, if the urine is stored under proper conditions (in darkness, in stoppered bottles containing glacial acetic acid, etc.) the amount of ascorbic acid lost over a period of twelve hours is only about ten per cent of the total; moreover, as the proportion so lost is fairly constant, the loss by oxidation should not, so long as the same test is employed, interfere with the compilation of standards of normality or the decision as to whether any particular patient has a low excretion of ascorbic acid.

More important, however, is the argument that 2:6 dichlorophenol-indophenol is reduced by certain other constituents of urine. Starting with Hopkins and Morgan (33), a number of workers have shown that the dye is reduced by such substances as glutathione, ergothionine, cystine and thiosulphate, of which the last two are normal constituents of urine. The original advocates of the dye test (20) claimed that the amount of non-specific reduction is equivalent to a maximum of 3 - 6 mg. out of a total of 20 mg./

20 mg. daily for a normal adult, and that such low values will not materially affect the comparison of results; but a consideration of the variations in the urinary excretion of cystine and thiosulphate hardly substantiates these claims. For cystine, Looney (34), using his colorimetric method, estimated the output of normal persons at 0 - 10 mg. per 100 cc. of urine (i.e. 0 - 150 mg. daily), and Medes (35), employing a different method, calculated that the normal limits for the daily excretion of cystine were between 1 and 84 mg.; moreover, in addition to normal individuals, with their very wide variations of excretion, there is also a rare - and frequently " missed" - disease of cystinuria, in which the daily output of cystine may be as much as 500 mg. (24), or about 33 mg./ 100 cc. urine. As for thiosulphate, the urinary excretion of this subst--ance depends on the amount of protein eaten: consequently a high protein diet may cause an increased decolorisation of the dye (36 & 37).

These points militate against the specificity of the dichlorophenol-indophenol test as applied to apparently healthy persons, and help to explain why such workers as Ingalls (38), Hawley et alii (39), van Eekelen (10),Youmans and others (40), Parsons (41), and Wright and his co-workers (42) have found such marked variation in the excretion of individuals on the same diet, and even in the excretion of the same person at different times, although Wright's sug--gestion that the differences are due to individual variations in the absorption of vitamin C is by no means to be lightly discarded. These points may also afford a clue to the hitherto unsolved riddle of why some cases of frank scurvy appeared to excrete up to 20 mg. of ascorbic acid daily. (22).

Even if the dye test be deemed sufficiently specific for application to healthy persons, any attempt to apply it to disease raises further possible fallacies: patients suffering from diseases commonly receive drugs, and we do not yet know all the drugs which affect the excretion of ascorbic acid. We do know, however, that if the urinary pH is raised by giving sodium bicarbonate the excretion of vitamin C is reduced (possibly through destruction of the vitamin in the alkaline bladder), whilst a reduction of the pH by the administration of ammonium chloride increases the excretion of the vitamin (43 & 44); and that treatment with salicylates, including aspirin, also raises the output of vitamin C.(45).

The dye test, with its many fallacies, is only a very approximate indication of the actual level of vitamin C excretion. Hence standards of " Normality " and of " Health " based solely on this test are unlikely to be particularly accurate. Of the various " Normals " and " Standards " that have been laid down, a few may be ment--ioned as examples: Harris and others (46), using a rather inadequate/

inadequate number of patients, assessed the normal daily concentration of vitamin C in urine at 0.03 mg. per cc., or for an average adult a total excretion of about 45 mg. daily; Harris and Ray (47), after careful investigation, concluded that a urinary concentration above .01 - .02 mg. per cc. implied adequate nutrition in respect of ascorbic acid, or, in terms of total excretion, that an adult whose daily output was above 15 - 30 mg. could be regarded as free from any hypovitaminosis C; Abbasy and his co-workers (20) suggested that the minimum daily excretion for a healthy adult should be 10 mg.; and Toverud (48) found that the average excretion of adult females was about 37 mg. per day.

These standards of very limited and partial accuracy are mentioned here only because - as will be shown later - certain conclusions regarding the excretion of vitamin C in infectious diseases have been based solely on the dye test.

(C) Saturation Tests.

The principle of giving a test dose of a substance and investigating its fate is well known in medicine, - e.g. the laevulose tolerance test for hepatic efficiency. Applying this principle to ascorbic acid, numerous workers have enabled us to form a clear idea of the general trend of results: when a large dose of vitamin C is given to an individual who is already well nourished in respect of the vitamin, he at once responds by an increased excretion of that substance; in other words, his tissues are more or less saturated, so that much of the test dose has to be excreted. On the other hand, poor nutrition in respect of the vitamin is revealed by the absence of any marked increase in the excretion; the tissues are not nearly sat--urated, and for a few days they absorb most of the addit--ional ascorbic acid administered.

Whilst the general picture is clear, the questions of the optimum dosage for the test and of the exact interpret--ation of results are still controversial.

The original propounders of the saturation test (46, 47 & 20) advocated an oral dose of 700 - 1,000 mg. of the vitamin daily. They said that all subjects free from hypovitaminosis respond by an abrupt rise in excretion, whereas " unsaturated " persons retain the excess vitamin C to remedy the unsaturation; and they further claimed that the point of demarcation lies in the vicinity of a pre-dosage excretion of 13 mg. per day. Using this test in conjunction with the ones previously mentioned, Harris and others (49) attempted to devise standards for the intake and output of the vitamin. They fed six adults on a diet containing little or no vitamin C until their reser--ves were very low, and then gave 25 mg. of the vitamin daily; the excretion rose steadily for six or seven weeks to/

to 13 - 14.8 mg., and at this point all six gave " good responses " to a test dose of 700 mg. In a subsequent experiment they gave 40 mg. per day, and after five weeks the excretion became stabilised at 25 - 29 mg. " On the basis of this work, an excretion of 13 mg. daily, for an average adult of ten stone weight, has been described as the minimum standard excretion. " (18)

For these suggested standards there is really very little evidence. The intake of 25 mg. daily was based mainly on Gothlin's "reputed minimum optimal dose" (50), which - being itself based on the wholly invalid test of capillary fragility - is valueless. The standard of above 13 mg. for the excretion has been criticised by Godber (18) on the ground that it merely indicates a state of saturation and that there is no evidence that unsaturation is incompatible with health. It might also be criticised on the ground that, since Harris accepted as satisfactory an excretion of 50% of the test dose, the "good responses" to a test dose, by individuals who were previously excreting 13 - 15 mg. per day, may indicate a considerable degree of unsaturation: and a degree of unsaturation so great that nearly half of the test dose is retained by the tissues may well be below the minimum compatible with perfect health. Hence these suggested standards - an intake of 25 mg. and an output of 13 mg.- may be either too high or too low. Moreover, these standards were constructed from a grossly inadequate number of cases, - six in all; and the investigator who originally claimed (in 1935) that 25 mg. represented the minimum adequate daily intake maintains in 1942 that over 30 mg. are necessary. (57)

Archer and Graham (51 & 52) regarded an excretion of half the test dose as unsatisfactory. Accordingly, they suggested a test which involved calculating the total amount of ascorbic acid given before the subject began to excrete 75% of his daily intake of 1,000 or 400 mg. On the other hand, Parsons (41) advocated the use of a higher test dose, with an excretion of 10% regarded as indicating saturation.

Ralli and his co-workers (53) and Finkle (54) pointed out that results from oral test doses might be partially vitiated by individual differences in degree of absorption. They therefore advised an intravenous test dose of 100 mg. Wright and others (42) accepted the principle of an intra--venous test dose, but preferred to give 1,000 mg.

Probably the technique most commonly used today is Harris's modification of his original method: a daily test dose of 70 mg. per stone of body weight is given, the vitamin C content of urine voided four to six hours after each dose (i.e. at the time of the peak of the excretory response) is estimated by titration of dichlorophenol-indophenol/

indophenol, and the degree of unsaturation is measured by counting the number of days before the curve of excretion rises to a plateau. Harris, who showed in 1940 (55) that, as the curve of excretion rises very steeply, the exact figure taken for the lower limit of saturation does not matter greatly, has recently claimed (56) that for persons with a high intake of vitamin C saturation is completed on the first day of dosing, for persons with just sufficient vitamin C in their diet the plateau is reached on the second day, failure to reach the plateau until the third day or later indicates deficiency of the vitamin, and in fully developed scurvy saturation is not completed for 7 - 10 days. In a subsequent article (57) he states, -

> "In my experience, an intake not greatly in excess of the League of Nations marginal standard (30 mg.) will suffice to keep the organism at such a level that saturation, as already defined, is reached on the second day of test-dosing."

Among the many workers who have used saturation tests may be mentioned Abbasy, Harris, and their colleagues (58, 59, 60 & 61), Jetter and Bumbalo (62), Gander and Niederberger (63), Emmerie and van Eekelen (64), van Eekelen and Heinemann (65), Widenbauer (66), Scarborough and Stewart (67), Guggenheim (68), and Francis and Wormall (69).

(D) Urinary Estimations and Saturation Tests, - Conclusions.

If the figures given separately by Medes and Looney for the excretion of cystine are correct, the ordinary dye test for the amount of ascorbic acid in the urine can be of but little value; and even if we accept Harris's claim that the non-specific reduction never amounts to more than 6 mg. per day, such non-specific reduction is enough to give a very erroneous idea of the vitamin C excretion. It might, for example, make a man whose real output was only twelve milligrammes appear to be excreting half as much again. Also, the collection and storage of all the urine passed is no easy matter: to say nothing of the loss of defaecation urine, an ambulant patient may visit the lavatory unnoticed, an ill patient may be slightly incon--tinent, or a nurse may inadvertently expose the stored urine to light or (by failure to replace the stopper) to air; any such event may reduce the apparent excretion by, say, 25 %. The alternative method of estimating merely the concentration of a single specimen has the disadvantage that the concentration cannot be relied on to remain stationary during the entire day. Moreover, an already wide margin of error is made even wider by the fact that the excretion of vitamin C varies with the pH of the urine. The ordinary urine test, then, is a very rough approxim--ation of limited value.

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The saturation tests, on the other hand, are fairly accurate: when we are dealing with a dramatic rise in the excretion of vitamin C (e.g. from a pre-dosage level of 20 mg. to a height of 200 mg.) a non-specific reduction accounting for a few milligrammes becomes of negligible importance; moreover, as was shown on page 7, we do not need to collect the entire day's output of urine, since the concentration at the period of maximal response is all that is required.

The great difficulty with the saturation tests is that, since administration of vitamin C may have a thera--peutic effect, they cannot be used in the initial construction of standards. For example, prodromal symptoms of scurvy have been noted since the days of Admiral Sir John Hawkins (who described them in 1569), but, if we are confronted with a case that resembles a prodromal scurvy, we cannot use a saturation method to ascertain what extent of hypovitaminosis C exists in prodromal scurvy, because the ascorbic acid would prevent the scurvy from developing, and so leave us uncertain whether our suspect had really been a case of pre-clinical scurvy.

Once definite standards are laid down, saturation tests are of real value; but in the past conclusions from such tests have sometimes been based on arbitrary and unjustified standards.

(E) The Intradermal Test.

Rotter (70), Portnoy and Wilkinson (71) and others have used an intradermal test for the estimation of nutritional status in respect of vitamin C. A sterile solution is prepared, containing 2 mg. of 2:6 dichlorophenol -indophenol dissolved in 4.9 cc. of distilled water; from a finely graduated syringe .01 cc. of the solution is injected intradermally, raising a wheal of about 2 mm. diameter; and the time required for the complete disap--pearance of the blue colour is noted. The proof that the disappearance of the colour is due to reduction of the dye, not to mere absorption from the area, is supplied by the fact that on intradermal injection of other dyes (e.g. methylene blue) no similar decolorisation occurs.

It is difficult to inject exactly 0.01 cc., but error from this source is minimised by injecting approximately 0.01 cc. into four separate spots on the patient's arm and taking the average time required for the disappearance of the colour.

Portnoy and Wilkinson, who found that the results of the intradermal test correlated well with blood estimations, claimed/

claimed that " Individuals who are on a 'good' diet with regard to the vitamin give decolorisation times of of less than ten minutes, while those taking large amounts of vitamin C in their food give decoloris--ation times below five minutes. On the other hand, prolongation of the decolorisation time over ten minutes is suggestive of a lack of vitamin C in the tissues. " (72)

Although certain other workers (73) have contended that the test was unsatisfactory, it appears to give a reasonably accurate indication of the general state of the body in respect of ascorbic acid. However, as it has not been used in investigations of infectious patients, the intradermal test need not be further considered.

(F) Quantitative Estimates of Vitamin C in Plasma and Blood.

Despite the relative popularity of urine tests, it is obvious that questions of urinary excretion and of renal threshold are not fundamental: what really matters is the nutritional state of the tissues. Hence examination of the blood should be the most direct and reliable method of determining vitamin C requirements and standards.

As early as 1934 van Eekelen and others (74) devised, for the estimation of plasma ascorbic acid, a method which involved preliminary precipitation of the proteins with trichloroacetic acid. This method, however, was later superceded by the method of Farmer and Abt (75), which, as originally described, involves the preparation of a tungstic acid, protein-free filtrate, and titration of this with a solution of dichlorobenzone-indophenol: 5 cc. of blood are withdrawn from a vein, oxalated, and centrifuged; the supernatant plasma is pipetted into a large centrifuge tube, to which there are added 4 cc. of distilled water, 2 cc. of 5% sodium tungstate, and - after thorough mixing of the previous contents - 2 cc. of one third normal sulphuric acid; after standing for two minutes the tube is centrifuged, and a clear fluid obtained; the fluid is then immediately titrated against a previously standardised solution of sodium 2:6 dichlorobenzone-indophenol. This method gives the reduced ascorbic acid content of the plasma.

A method of estimating the vitamin C content of whole blood was devised by Mirsky and his co-workers (76): 5 cc. of oxalated blood and 5 cc. of 10% trichloroacetic acid are mixed, and added to 5 cc. of N/6 mercuric acetate; .23 Gm. of calcium carbonate are added and the mixture is stirred thoroughly; the solution is then centrifuged, and a clear fluid obtained; to regenerate reduced ascorbic acid, H_2S is bubbled through, and excess of that gas is removed with nitrogen; finally the clear fluid is titrated against dichlorophenol-indophenol/

dichlorophenol-indophenol.

A modified technique for the estimation of vitamin C in plasma was suggested by Pijoan and Klemperer (77). An alternative method for whole blood was devised by Lund and others (78), employing methylene blue as an indicator. And Goth (79) used a saturation test: he determined the plasma level of ascorbic acid before, and two hours after, an intravenous injection of 300 mg. of the vitamin; and he claimed that, if the second concentration was found to be less than twice the first, a state of hypovitaminosis C existed.

From the work of these investigators and of others who have studied the ascorbic acid content of blood or of plasma - e.g. Abt and Farmer (80) some three years after their original description of their method, Bajardi and Margulius (81), Faulkner and Taylor (82), Rhinehart and others (83), Wortis and his co-workers (84), Goldsmith and Ellinger (85), etc. - two main points emerge:-

(a) There appears to be no real difference between plasma concentration and concentration in blood; in other words, the vitamin is evenly distributed between the plasma and the corpuscles.

(b) From the very considerable amount of published work it should be possible to derive definite standards for the amount of ascorbic acid in human blood.

Before the standards for the level of vitamin C in blood and plasma are considered, it should be noted that there are two possible fallacies in the plasma tests: a low plasma concentration of ascorbic acid may occur in a person who has adequate reserves in his tissues, purely as an immediate result of a reduction in his intake of the vitamin (e.g. a febrile patient receiving a milk diet with little vitamin C); and a high plasma concentration, while usually indicating an adequate store in the tissues, might be merely an immediate and temporary response to a large dose of the vitamin.

(G) Levels of Ascorbic Acid in Blood and Plasma.

The fallacies of the direct urine test and the impos--sibility of constructing initial standards from the satur--ation test were demonstrated in an earlier section; and any lingering feeling that the dye test of urinary excretion could indicate the general state of the body in regard to vitamin C should be dispelled by the findings of Abt, Farmer and Epstein (11), and of van Eekelen (10): the first named group of workers showed that, while the amount of vitamin C in the plasma is closely correlated with the intake, there is no correlation between the concentrations of the vitamin in the plasma and in the urine; and van Eekelen, maintaining

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a young adult an a diet devoid of vitamin C for 83 days, found that, although the plasma concentration fell quite steadily from 1.7 mg./100 cc. to 0.19 mg., the urinary excretion fell only from 13 mg. daily to 7 mg. (In other words, either the dye test was inaccurate or the patient had no renal threshold for the vitamin).

In the blood ascorbic acid is approximately evenly distributed between the plasma and the corpuscles (86). In investigating the different "Normal" findings of various workers, we have to remember that the results can be expressed in three ways: (a) the vitamin C content of whole blood can be ascertained and expressed as mg./100 cc. of blood; (b) the vitamin C content of plasma can be calculated and expressed as mg./100 cc. of plasma, - giving a figure similar to that of (a); and (c) the vitamin C content of plasma can be ascertained and expressed as mg./ 100 cc. of the blood from which the plasma was derived, - giving a figure about half of that in (a) and (b). A hypothetical example will make this clear:- 100 cc. of blood contain approximately 50 cc. of plasma (87); if the 50 cc. of corpuscles contain 0.5 mg. of vitamin C and the 50 cc. of plasma contain 0.5 mg. of the vitamin, then it can be said (a) that the concentration of vitamin C in the blood is 1 mg./100 cc. blood, or (b) that the concentration in the plasma is 1 mg./100 cc. plasma, or (c) that the concentration in the plasma is 0.5 mg./ 100 cc. of blood. This point of terminology has to be watched.

Broadly speaking, most workers have found that the average ascorbic acid content of blood or plasma - expressed as in (a) or (b), above, not as in (c) - is about 0.5 - 0.7 mg. /100 cc., with fairly wide individual variations: thus Rhinehart and others (88) calculated that the plasma levels of medical students, a well nourished section of the com--munity, varied between 0.25 and 1.45 mg./100 cc., with the average about 0.75 mg.; Abt and his colleagues (11), investigating young adults, found that all their cases fell between 0.109 and 0.735 mg./100 cc., but in a subsequent experiment (75) they calculated that the plasma range of concentrations in normal people was much higher, namely, from 0.687 to 2.29 mg./100 cc.; Portnoy and Wilkinson (72) regarded 0.6 to 1.85 mg./100 cc. as normal, and figures of below 0.45 mg. as indicating a dangerous depletion of the body stores; Schroeder (89) took 1 mg./100 cc. as the normal blood level (and 50 mg. as the minimum daily intake); Ingalls (38) tried to formulate standards of plasma vitamin C thus -

Optimum	Saturation,	2. - 1. mg./100 cc.
	Normal,	1. - .7 " "
	Low,	.7 - .5 " "
Suboptimum,	- - - -	.5 - .3 " "
Asymptomatic scurvy,		.3 - .15 " "
Scurvy,	- - - - - - - -	.15- 0 " "

Eddy and Dalldorf (90), in an authoritative exposition, said " In our experience blood assay has yielded values of nearly 1 mg./

1 mg. per cent in the vast majority of adults "; and a leading article in the 'British Medical Journal' of 14th. February, 1942, quotes Minot's figure (91) of 0.7 mg. per 100 cc. of plasma as the minimum concentration indicating a satisfactory nutritional state in respect of vitamin C.

Standing foursquare against all these findings are the anomalous results of Fox and his co-workers. (73). For seven months they investigated two parallel groups of natives employed in one of the gold mines of the Witwater--srand, each group consisting of 950 individuals. Group A received the " usual diet ", containing about 15 -25 mg. of ascorbic acid daily, whilst Group B received an addit--ional daily ration of 40 mg. of vitamin C per person, in the form of an orange-juice concentrate. In Group A 12 persons developed scurvy, as compared with only one case in Group B, but no significant difference was found between the groups in regard to general condition, gain or loss of weight, working efficiency, susceptibility to infections (e.g. influenza, pneumonia, varicella, and tuberculosis), or rate of healing of wounds and fractures. The ascorbic acid content of the plasma was estimated in 74 healthy members of Group A, 86 similar members of Group B, and 17 cases of scurvy (including a few not in the original experiment). Fox and his colleagues record the levels of plasma vitamin C in mg. per 100 cc. blood: to avoid confusion (see page 11) their figures are therefore here converted into the more usual terminology. Their findings may be summarised thus:-

	Vitamin C in mg./100 cc of plasma.	
	Average.	Range.
74 controls (after seven months)	0.46 mg.	0.30 - 0.80.
86 men given extra vitamin C (after seven months)	0.504 mg.	0.28 - 0.96.
17 cases of scurvy	0.348 mg.	0.20 - 1.0.

Also, the capillary fragility test was negative in every patient tested, Dalldorf's method being the one employed; and Rotter's intradermal test gave practically no differ--ences between test cases, healthy controls, and cases of scurvy.

From these results Fox and his co-workers conclude that the daily vitamin C requirement of native mine lab--ourers, even when engaged in strenuous physical exercise, is very small indeed, and that " A net daily intake of about 15 mg. of ascorbic acid, perhaps even less, can evidently protect the majority of such individuals from scurvy." They would, presumably, also maintain that a plasma concentration of 0.46 mg./100 cc. is shown to be compatible with health.

After a detailed study of their paper, the present writer is disposed to draw very different conclusions:- The 950 controls received a diet containing 15 - 25 mg.of vitamin/

vitamin C daily, and it may be that some of them devised methods of supplementing their diet. Of these 950, twelve developed clinical scurvy, and tests of physical efficiency showed that the whole group was in poor condition: e.g. in putting a 16 lb. shot, out of 107 men tested not one achieved even the comparatively poor throw of thirty feet. Hence it appears that an intake of 15 - 25 mg. and a plasma concent--ration of about 0.46 mg/100 cc. are below the level at which scurvy is excluded, and are also below the minimum compatible with good physical condition. As for the 950 individuals who received an additional 40 mg. of vitamin C each day, although even the very slight amount of absorption that took place (a rise in the average plasma concentration from 0.46 to 0.50 mg %) appears to have made a reduction of statistical significance in the incidence of scurvy, the failure of the plasma levels to rise materially in response to prolonged administration of the vitamin makes it manifest that most of the additional ration was either not absorbed or not utilised. This failure of absorption or utilisation explains why the intradermal test revealed no differences between the groups; and it makes the comparison of the low standards of physical efficiency of Group A with the equally low standards of Group B quite pointless.

In explanation of this failure of absorption or of utilisation one can only suggest tentatively that deficiency of some other substance may limit the absorption of vitamin C. - In respect of this conjecture the work of Kasahara and others (92), on the relationship between vitamins B and C, is interesting, although not sufficiently relevant for discussion here.

The findings of Fox and his co-workers have been con--sidered in some detail, because their conclusions, if they were justified, would contradict the view of practically all recent investigators, - that plasma concentrations of below 0.5 mg. per 100 cc. are both uncommon and dangerous.

(H) Standards of Ascorbic Acid. - Some Reflections and Conclusions.

It is sometimes insufficiently realised that there is a vast difference between the positive condition of health and the negative state of absence of disease; between the energy and joie de vivre of excellent physical condition, with optimum nutrition, exercise, rest, etc., and the state of a man who, although suffering from no organic disease, carries out his daily work with an effort which leaves him listless and apathetic. In the medical profession alone there must be many today who, though they regard themselves as normal, " healthy " individuals, find that - whether sapped by overwork or by rationing or by nervous strain - their powers of endurance, their emotional stability, and even their ability to concentrate are far less in 1942 than in/

in the days of peace.

Health, then, is not simply the negation of disease.

Since anorexia, lowered physical endurance, listless--ness, and undue fatigue have for centuries been recognised as among the prodromal features of scurvy - " Some show it by their lasinesse ", wrote Hawkins in 1569 - it does not seem unreasonable to suggest that there may exist a state in which the body contains enough ascorbic acid to prevent the development of clinical scurvy, but not sufficient to prevent the appearance of some of the prodromata: or that, in other words, a relative lack of vitamin C (a defic--iency not great enough to cause obvious disease) may be one of the many factors which can lower human vitality and energy. To summarise the steadily accumulating mass of evidence that has already converted this hypothesis into proved fact does not fall within the scope of this essay: but a few examples may be cited:-

Gander and Niederberger (93), administering additional ascorbic acid to elderly persons whose ascorbic acid levels had previously been low, recorded increased vitality, improved general health, and better sleep among the results. Stephenson, Penton and Korenchevsky (94) in 1941 studied the effects of administration of vitamins B and C, separ--ately and simultaneously, to elderly people, using 40 senile patients for their experiment and 18 others as controls: their very modest conclusion, that prevention of latent or partial vitamin deficiencies would probably result in the approach of a less pathological senility, is more than confirmed by their results, a few of which can be summarised as follows:

	After a course of vitamin C. Number of cases in which the senile feature was			After a course of vitamins B & C. No. of cases in which the senile feature was		
	Better.	Worse.	Unchanged.	Better.	Worse.	Unchanged
Shuffling gait	16	0	9	19	0	5
Uncertain movements	12	1	9	17	0	4
Impaired mentality	13	3	24	25	3	11
Oedematous eye sacs	21	1	10	25	2	5
Skin rashes	6	0	7	10	2	5
Insomnia	12	2	3	14	1	1

In 1942 Glazebrook and Thomson (95) found that the giving of additional vitamin C to 300 lads of 15 - 20 years considerably reduced the duration of tonsillitis, and that of these 300 boys not a single one developed rheumatism or pneumonia while receiving the supplement, whereas in the same/

same number of controls there were half a dozen cases of each disease; and, some two years earlier, Roff and Glaze--brook (96) showed that in British naval trainees the incidence of gingivitis in those given supplementary vit--amin C was only one quarter of that in trainees receiving no such supplement.

In Germany (97) more than one and a half million children of 10 - 14 years were given a daily supplement of 50 mg. of synthetic ascorbic acid. This was found to result in an acceleration of physical development and an increased resistance to infection.

Hirata and Suzuki (98) claimed that administration of vitamin C improved patients suffering from progressive muscular atrophy; Lyle and McLean (99) showed that intra--venous injections of ascorbic acid caused a dramatic improvement in cases of punctate and phlyctenular keratitis; Hunt (100) demonstrated that the healing of a wound is pro--foundly disturbed if the vitamin C content of the body is low, a finding that has recently been confirmed, in respect of dental extractions, by other workers (101); and Crandon and Lund (102) have corroborated that, in volunteers living on a scurvy-producing diet, lassitude and fatigue appear long before any objective signs of scurvy.

Despite a few sceptics like Rietschel (103) - who anticipated Crandon in voluntarily starving himself of vitamin C, and denied the existence of any ill effects at the end of a hundred days of this starvation, although his plasma concentration of the vitamin was practically nil - there is a great, and steadily growing, mass of published work which supports the view that (in the words of a recent leading article in 'The Lancet') " There is a wide margin between the amount of vitamin C necessary to prevent scurvy .... and the amount necessary for a state of well-being. " (104)

We must look, then, for three standards of comparison for vitamin C: - the minimal requirement, below which a person is in danger of developing scurvy; the optimal requirement, or minimum amount compatible with perfect health and physical fitness; and, somewhere between these two, the "normal" or "average" findings among the popul--ation at large. Is it possible to determine these figures, even approximately ?

The minimal requirement - i.e. the amount necessary to obviate the danger of actual scurvy - is fairly easy to determine: it has already been shown (on page 14) that a daily intake of 25 mg. and a plasma level of 0.46 mg./ 100 cc. permit the occurrence of an occasional case of scurvy, and are therefore just below the minimum. In accepting Neuweiler's suggested minimum (105) of 30 - 35 mg. per day for an adult, or roughly 0.5 mg. per kilo, we have the support/

support of most of the leading authorities on the subject: e.g. the League of Nations' Technical Commission on Nutrit--ion advocated a minimum intake of 30 mg.(106); Harris has found that an intake slightly higher than the League of Nations' marginal standard will just reach the minimum standard of adequacy as determined by his saturation test (57); Scheunert (107) advises 30 - 50 mg. per day; and Davidson and Scarborough (86) assert that " The daily min--imum requirements of the normal adult male are in the neighbourhood of 30 mg."

The suggested figure of 30 - 35 mg. daily corresponds with a blood concentration of 0.5 - 0.6 mg./100 cc.

These generally accepted minima - an intake of about 0.5 mg. per kilo and a blood level of 0.5 - 0.6 mg./100cc. - are the amounts necessary to prevent scurvy, not, as some writers (108) have tried to maintain, the " amount necessary for health ".

It is to be noted, incidentally, that even these minimum figures are a little higher than the standard of adequacy originally suggested by Harris and others in 1936. This is important, because the old Harris standard has been employed in some of the work on vitamin C in relation to infections.

"Normal" or "average" figures are bound to vary with the country, the season, etc. Perhaps the best estimate for Britain is that of Widdowson and Alington (109), who investigated the diets of 63 women in time of peace -1935 - and of 57 women in wartime - 1941 -, and calculated that their average intake of vitamin C in 1935 was 57 mg. and in 1941 26.9 mg. per day. The last figure is just below the "minimum" level, but this can be correlated with the fact that cases of scurvy are now less infrequent than in pre-war days: e.g. two Edinburgh physicians have seen twenty-four cases of fully developed scurvy within two years, and Paterson and Daynes have recently commented on the increase in scurvy since the outbreak of war. (110 & 111).

The peacetime average of 57 mg. daily is, however, probably a little on the high side: for one thing,most of the women investigated were socially of the middle class. Perhaps a daily intake of 40 - 50 mg. would be a fairer average for normal times.

In considering the highly controversial question of optimum standards, the present writer proposes to start with a simple point that appears to have been overlooked in the past. - Among the many fluids of which the vitamin C concentration has been determined is human milk (112, 113 & 114). Remembering

> " The marvellous way in which the cells of the mammary gland pick out, from the salts of the circulating/

circulating plasma, exactly these salts which are needed for the growing animal, and in the same pro--portions " (115),

and remembering that the mother's diet, age, and condition of health are - in the words of Sir Robert Hutchison -

" almost entirely devoid of any appreciable effect on the composition of the milk " (117),

are we not entitled to conclude that the high level of the vitamin in the secreted milk, a level at least twice as high as that in the plasma of most mothers, is - to speak teleologically - an indication of Nature's decision regard--ing the optimum requirements of the infant ?

Using the previously mentioned calculations regarding the concentration of vitamin C in human milk, and using the figures given by Hutchison and Mottram (118)for an infant's requirements of milk, figures which themselves tally closely with those quoted by other recognised auth--orities (116), we can draw up a table as follows:-

Age of child.	Milk required, in cc.	Total vitamin C, (at 2 mg./100 cc. milk.)
2 weeks	533 cc.	10.67 mg.
4 "	633 cc.	12.67 mg.
8 "	780 cc.	15.6 mg.
12 "	800 cc.	16. mg.
16 "	866 cc.	17.33 mg.

Lack of definite knowledge as to whether there is any diminution in the ascorbic acid concentration of milk in the later months makes it unjustifiable to continue the table further. It appears, however, that in the early months of life nature provides the infant with over 3 mg. per kilo of body weight.

Estimates of the optimum requirements of vitamin C vary enormously. Perhaps the lowest is that of Kellie and Silva (119), who - in 1939 - contended that a daily intake of 15 mg. was sufficient to maintain normal indiv--iduals in good health, - an intake which would give an adult less than nature offers to a child of eight weeks, and which (far from being optimum) even falls below the minimum figure necessary to prevent scurvy. At the opposite end of the scale are the suggestions of Stepp and Schroeder (120), who claimed that the optimum intake for an adult was 300 mg. daily, and the figures of Everson (121), who advocated 7.5 mg./kilo for children of 3 - 5 years.

However, the great bulk of informed opinion appears to be converging towards optimum figures of 10 - 20 mg. daily for an infant and about five times that for an adult. Thus Smith (122) in 1938, after a careful review of the literature, suggested that 50 -60 mg. for adults, 40 for children, and 20 for infants " may be considered the middle or barely adequate consumption level with but little margin of/

of safety or allowance for individual variations in requirement". Matthews and Bacharach (123) claimed, in 1941, that a young infant required 5 to 10 mg. per day. Holmes and others (124), in 1941, accepted 50 mg, daily as the requirement of an average adult. Heinemann (37) maintained that an adult needed 60 mg. Sweeney and his co-workers (125) in 1941 advocated 50 - 60 mg. daily in health and 200 mg. daily in disease. In the same year, Davidson and Scarborough (86) in an authoritative review of vitamin C declared that the optimum dose for an adult was 60 mg. daily, and for an infant at least 15 mg. A leading article in the 'British Medical Journal' (126), in 1942, quoting the work of Minot and others in support of a satisfactory serum level of 0.7 mg./100 cc., says,-

> " The most generally accepted estimate of the daily requirement of vitamin C by the adult is 50 to 60 mg. daily, and much more in both chronic and acute illnesses."

The United States National Research Council (127) stated in 1941 that the desirable daily intake for an adult was 75 mg. And, finally, Youmans (128), in his new book on "Nutritional Deficiencies: Diagnosis and Treatment", says,

> "Recently studies based on newer biochemical tests to determine amounts needed to maintain a normal body store have indicated amounts considerably larger than those suggested in the past. On the basis of these studies a fair to good but not 'luxus' intake for infants would be 20 to 50 mg. daily, for children 40 to 100 mg., and for adults 50 to 100 mg. "

Approximate estimates of minimum, optimum and average levels may be tabulated thus:-

	Daily intake in mg.	Mg./kilo.	Plasma level in mg./100 cc.
1. Minimum to prevent danger of scurvy.			
Adult	30	0.43	0.5 - 0.6
Infant	5	1.5	
2. Optimum.			
Adult	60	0.86	0.8 - 1.
School child	40 - 60.	1.5 - 2.	1. - 2.
Pre-sch. "	25 - 40.	2. - 2.5	
Infant	10 - 25.	3. - 3.5	
3. Average (pre-war).			
Adult	40 - 50.	0.6 - 0.7	0.6 - 0.7

(Wherever there is insufficient evidence regarding plasma concentrations, the last column has been left blank.)

Until the Meiklejohn and Stewart method comes into general use exact correlation with excretion is impossible, but - on the basis of the dye test - it may be suggested that for adults the minimum intake corresponds very roughly with a daily excretion of 14 to 20 mg. (or a concentration of .01 to .015 mg./cc. of urine); the optimum intake implies an/

an excretion of round about 30 to 40 mg. per day (or a concentration of about .02 to .03 mg./cc. of urine); and the average adult excretion is probably somewhere in the neighbourhood of 20 to 30 mg. (or a concentration of about .015 to .02 mg./cc. of urine.)

(2) APPLICATION OF THESE STANDARDS TO INFECTIONS.

The fons et origo of much of the work on vitamin C in relation to infections is a single case of coryza: in 1935, while Abbasy and others (20) were investigating the excretion of patients on a standard diet, one of their subjects developed a mild coryzal fever, and his urinary excretion - as estimated by the dichlorophenol-indophenol test - fell from a steady level of 19 mg. to 9 mg. and remained there for a week. This event led the investig--ators to a study of the effects of pyrexia in general, and of various specific illnesses.

If the early workers on ascorbic acid had cared to dip into the records of the past, they would have been able to start their researches on the effect of infections without waiting for one of their subjects to catch a cold: in the days of sailing-ships many a mariner knew that scurvy was apt to follow in the wake of a wave of infect--ious disease. Numerous instances - too many to be due to mere coincidence - are on record of outbreaks of scurvy subsequent to epidemics.

That scurvy is due to deficiency of vitamin C is, despite the doubts expressed by Rietschel and Mensching (103) and Lauber and Bersin (129), too firmly established to require further proof. Scurvy as a sequel to acute infection implies that the acute infection causes some reduction in the bodily reserves of ascorbic acid. This reduction might be caused in one of three ways, - by excessive excretion, by increased metabolism, or by decreased intake or absorption.

As the amount of published work on vitamin C levels in infections is quite considerable, it is convenient to start by considering various diseases separately.

(a) Rheumatic Fever.

In 1936, Abbasy, Hill and Harris (58) tried to inves--tigate the excretion in rheumatic fever and tuberculosis, using the dye test and, subsequently, a saturation test; their experiment embraced 107 active cases of rheumatic fever, 86 convalescent cases of that disease, 88 cases of surgical tuberculosis, and 64 healthy children. As all the children investigated were in hospitals or other institutions, details of diet, etc., were under control. It/

It will be remembered (see page 7) that Harris had prev--iously suggested as a standard of adequacy an intake of 25 mg. for an adult and an excretion of 13 mg. or about 1 mg. for every 10 lbs. of body weight, - a standard which has been shown to be dangerously low. Using this low standard, the investigators found that the excretion of healthy children and of quiescent cases of surgical tuber--culosis was, without exception, " satisfactory "; that active rheumatic cases excreted an average of only 1 mg. per stone of body weight (i.e. only about two thirds of the low total then accepted as adequate); and that 83 % of the convalescent rheumatic patients had a " subnormal" excretion. Using the more reliable saturation test, they found that the rheumatic cases were grossly unsaturated: the test doses had to be repeated for 2 to 4 days before there was a significant increase in the urinary excretion of ascorbic acid; and the response of the controls was greater after a single test dose than that of the rheumatic cases after their third dose.

Subsequently (59 & 61), the same workers produced further evidence that the excretion of vitamin C was red--uced in rheumatism and tuberculosis, and that the output rose during convalescence and healing.

More important, because based on a more reliable means of investigation, is the work of Rhinehart and others (83 & 130) on plasma levels in patients suffering from active rheumatic fever. They found that the plasma concentration was low, and, since some cases responded slowly to large oral dosage, they suggested that absorption was defective. They also claimed (vide infra) that hypovitaminosis C was was of aetiological importance in rheumatic fever. Sendroy and Schultz (131), while refuting the claim that lack of vitamin C played a part in the causation of the disease, found some degree of apparent ascorbic acid deficiency in eight of their thirteen cases, even though their standards of " normality " were very low. Perry (132), after a study of the excretion of eleven rheumatic patients, concluded that, while " Vitamin C deficiency is not an important factor in the causation of acute rheumatism ", " mild degrees of this deficiency are not uncommon in rheumatic fever."

In contradistinction to all these workers, Parsons (41) and Keith and Hickmans (133) found an increased excretion of vitamin C in acute rheumatism, but their results may well have been due to the effect of salicylates.

The general trend of the evidence relating to ascorbic acid in acute rheumatism is to show that the bodily reserves are depleted, and that - since the urinary excretion is also reduced - this depletion is not the result of an increased urinary/

urinary output. The depletion may be due to one or more of three factors: there may be defective absorption, as a result of digestive disturbance; there may be an increase in the metabolism of ascorbic acid; and there may be an increased excretion of the vitamin by a route other than the urinary one. In connection with the last of these possibilities, it is pertinent to recall that Harris (57) has recently found that vitamin C is lost in measurable amount in the sweat, and that profuse perspiration is one of the features of acute rheumatism.

(b) Tuberculosis.

In 1936 Hasselbach (134) and Heise and Martin (135) independently produced evidence of gross vitamin C unsaturation in pulmonary tuberculosis. The last named workers subsequently claimed (136) that the urinary excretion of the vitamin was inversely proportional to the severity of the disease. The published work of Heise and Martin certainly indicates that a considerable degree of hypovitaminosis C existed in their tuberculous patients: of 106 tuberculous persons tested, the output of only 16 reached the very low standard of 14 mg. daily; 45 excreted between 8 and 14 mg.; and the remaining 45 excreted less than 8 mg. Despite the various fallacies inherent in the dye test, these findings afford definite evidence of deficiency. The claim that the excretion of the vitamin was inversely proportional to the severity of the disease does not, however, appear to be justified by the published figures: approximately one third of active advanced cases and one third of active moderate cases excreted less than 8 mg. per day; and a comparison of 42 active cases of moderate or advanced tuberculosis with 52 similar inactive cases shows that the latter ~~excreted~~ excreted rather less vitamin C than did the active cases.

The same workers, this time in collaboration with Schwartz (137), brought forward more evidence of the extent of hypovitaminosis C in tuberculosis. Their conclusions are reinforced by the results of Hurford (138) and by those of Abbasy, Harris and Ellman (61). These last investigators found that, although all their cases had received a daily supplement of ascorbic acid for four weeks prior to the beginning of the experiment, the daily excretion of 19 active cases was only 5 to 13 mg., that of 6 quiescent cases was 18 to 23 mg., and that of 13 "moderate" cases was intermediate between the other groups. Abbasy and his co-workers also found a correlation between the severity of the infection, as shown by the blood sedimentation rate, and the excretion of the vitamin, -i.e. the more severe the infection the less the excretion.

Views based solely on urinary excretion might well be suspect, but there is ample evidence based on blood examinations/

examinations: thus Dagulf (139) in 1939, examining 326 healthy persons and 255 tuberculous persons, showed that - even though the ascorbic acid concentration of his supposedly " healthy " group was a little below what is usually regarded as normal - the average concentration of vitamin C in the blood of the tuberculous patients was almost exactly one half of the average for the healthy people. Again, the Groth-Petersens (140), using the Lund-Lieck methylene blue method, and examining the blood of 260 tuberculous patients, concluded that such patients require a greater amount of ascorbic acid than do healthy people, if the serum concentration of ascorbic acid is to be maintained at the same level. Sweeney (125), too, in 1941, found - on the basis of blood and urine examinations of a large number of tuberculous patients - that moderate tuberculosis was associated with mild hypovitaminosis C, and that marked deficiency of the vitamin occurred in advanced tuberculosis.

Lastly it should be mentioned that in 1940 certain Liverpool workers (141), estimating the urinary excretion of 13 tuberculous patients prior to the therapeutic admin--istration of vitamin C, not only corroborated the usual finding of diminished excretion, but also re-affirmed that the amount of the ascorbic acid excretion was, roughly, inversely proportional to the severity of the infection. While this last assertion may or may not be generally valid, it does seem that in pulmonary tuberculosis the existence of hypovitaminosis C is definitely proved.

(c) Pneumonia.

As early as 1935 Schroeder (142) commented on the high degree of vitamin C deficiency commonly encountered after pneumonia; and this finding has been confirmed by Guldager and Poulsen (143), by Harde and his co-workers (144), and by various later investigators. In particular, Gander and Niederberger (63) have made hypovitaminosis C part of the rationale of their treatment of pneumonia: but this will be considered in a later section.

(d) Pharyngitis.

In an article with the provocative title, " Ist die Pharyngitis eine C- Hypovitaminose ? ", another Swiss worker (142) has told how, in a patient suffering from severe pharyngitis, he found a vitamin C deficiency so gross as to suggest that inflammation of the pharynx might be the first symptom of scurvy, and of how the pharyngitis cleared up without any treatment except oral and parenteral administration of vitamin C. This led him to investigate thirty-five subsequent cases of pharyngitis, in every one of which he found a very marked deficiency of ascorbic acid.

(e) /

(e) Other conditions.

For other infections the great bulk of the published work is in accord with the results already cited for acute rheumatism, tuberculosis, pneumonia and pharyngitis. Thus Woringer and Sala are quoted by Ormerod and Unkauf (146) as having observed four cases of scurvy precipitated by whooping cough; and Ormerod and Unkauf themselves found that the excretion of vitamin C in cases of whooping cough dropped almost to zero. Bamberger and Wendt (147) claimed that even large doses of ascorbic acid failed to increase the urinary output of the vitamin in cases of severe diph--theria. Kundiger and Salus (148) confirmed the existence of hypovitaminosis C in diphtheria; and Ghosh (149), Lueg and Hammann (150), and others have added their quota to the accumulating mass of evidence. Many of these workers also tried therapeutic administration of the vitamin, and must therefore be considered in a later section.

That hypovitaminosis C exists in diphtheria and in scarlet fever was denied by Godber (18), whose findings will be discussed presently.

As for pyrexia alone, Falke (151) carried out a very elaborate experiment just before the outbreak of war: measuring the concentration of ascorbic acid in the blood of 15 patients during febrile and afebrile periods, he tried to determine the exact amount of the vitamin necessary to prevent a fall in the blood level during the febrile periods. Falke concluded that approximately a hundred milligrammes more of vitamin C were required daily in the presence of fever than in its absence. It is to be noted that Falke's work does not imply that in pyrexia an additional 100 mg. of the vitamin are metabolised daily: not all of the supple--ment is absorbed from the alimentary tract.

(f) Anomalous Findings.

In the last few pages twenty-nine separate pieces of evidence have been cited in support of what Davidson and Scarborough (110) call " The well-recognised finding that acute and chronic infections .... are associated with a low ascorbic acid content of the blood and urine." Against these findings stand the conclusions of Parsons, of Keith and Hickmans, and of Godber.

An overwhelming preponderance of experimental results is really enough to decide a case, but the verdict can be given more confidently if the few results not in harmony with that verdict can be explained. It seems worth while, then, to consider in some detail the evidence that has been adduced against the view that infection implies a diminution in the ascorbic acid level of blood and urine.

Parsons/

Parsons (41) studied the excretion of 103 children, divided into five groups: (a) 18 children suffering from acute rheumatic fever; (b) 12 children with other acute infections; (c) 24 surgical cases without any infection; (d) 31 children convalescent after rheumatic fever; and (e) 18 convalescent after other diseases. He found that the majority of the patients in Group (a) excreted more ascorbic acid than those of Groups (b) and (c); and that the majority of the rheumatic convalescents - Group (d) - excreted more than the convalescents of Group (e). He used no blood or plasma tests, and the many fallacies of the dichlorophenol-indophenol test for urinary vitamin C have already been indicated: for example, as his numbers were small, the accidental inclusion of an undetected cystinuric in either Group (a) or Group (d) might distort the average appreciably. Again, working at a time when the low standard of adequacy which Harris had originally suggested was still in vogue, Parsons might easily have included in Groups (c) and (e) a number of children who - by modern standards - were suffering from a considerable degree of vitamin C deficiency. But the most obvious explanation of Parson's findings lies in the fact that acute rheumatism is universally treated with salicylates, which increase the urinary excretion of ascorbic acid: hence the apparent high excretion of Group (a); and the output of Group (d) may have been estimated before the effect of the salicylates had passed off.

The same arguments apply to the results of Keith and Hickmans (133), who also dealt exclusively with rheumatism, and who also employed only the unsatisfactory urine test.

In his M. D. thesis on " The Excretion of Vitamin C in certain Infectious Diseases and in Pyrexia ", Godber (18) gave a very lucid summary of the relevant literature up to 1937 (including brief references to the work of Parsons and of Keith and Hickmans, which in both cases was first published in January of 1938), and then recorded in some detail his own findings with regard to the urinary excretion of vitamin C - as estimated by the dye test - in 101 infectious cases immediately after their admission to hospital, and in the same patients when convalescent.

Godber's main results can be rearranged and tabulated under two heads, - first, the urinary concentration of the vitamin in mg/cc., and second, for patients whose total output of urine was measured, the total excretion " on an adult basis." - A brief preliminary explanation of this second heading is perhaps desirable: direct comparison of the total output of children of varying ages is obviously useless; therefore, for comparative purposes, the excretion of each child has been converted (by the use of Godber's table of standard excretions) to that of an adult in a similar state of vitamin C nutrition: e.g. a child of five years /

years is shown by the table to excrete 25 % of the vitamin C excretion of a similarly nourished adult; hence, if the actual output of a boy of five years were 5 mg., he would be credited with an adult value of 20 mg. (N.B. By the standards of later workers, Godber's values for the output of young children, as compared with adults in a similar nutritional state, are unduly low; hence the standardised excretions will tend to be too high.)

Tabulated summaries of Godber's findings are given below, the following abbreviations being used to save space: S.F. for Scarlet Fever, Dip. for Diphtheria, P.P. for Puerperal Pyrexia, Ton. for Tonsillitis, and O.D. for Other Diseases.

(1) Concentration of vitamin C in mg./cc. urine.

Concentration in mg./cc.	No. of acute cases						No. of convalescents					
	S.F.	Dip.	P.P.	Ton.	O.D.	Total	S.F.	Dip.	P.P.	Ton.	O.D.	Total.
Above .03	5	10	0	2	0	17	0	1	0	0	0	1
.021 -.03	9	5	1	2	3	20	3	0	0	0	0	3
.015 -.02	8	9	0	1	4	22	3	6	0	1	0	10
.01 -.014	14	7	4	0	0	25	9	10	0	3	2	24
Below .01	7	2	1	1	1	12	23	9	0	1	2	35
No.tested	43	33	6	6	8	96	38	26	0	5	4	73
No. notexamd	5	0	0	0	0	5	10	7	6	1	4	28
Grand total	48	33	6	6	8	101	48	33	6	6	8	101.

S.F. cases:- Acute stage - Mean concentration, .016mg./cc;Median, .015
Convalescents - " " , .009 " ; " , .008

Dip. cases:- Acute stage - " " , .035 " ; " , .019
Convalescents - " " , .012 " ; " , .0105

No correlation noted between urinary concentration and either severity of case or degree of pyrexia.

(2) Total excretion of vitamin C on an adult basis.

Standardised Excretion of	No. of acute cases						No. of convalescents.				
	S.F.	Dip.	P.P.	Ton.	O.D.	Total.	S.F.	Dip.	Ton.	O.D.	Total.
Above 19.5mg.	8	9	0	2	1	20	2	2	0	0	4
13. - 19.5	8	8	2	3	1	22	11	5	2	1	19
Below 13.	8	3	4	0	0	15	16	8	1	0	25
Totals	24	20	6	5	2	57	29	15	3	1	48.

Since the diet of convalescents included green vegetables, potatoes and fresh fruit daily, tomatoes frequently, and oranges at least every second day, Godber argues that, after "at least a fortnight" of this diet, they can be regarded dietetically as normal people; and since the excretion of the patients during the acute stage was higher than that of the "normal /

" normal people", he concludes that the urinary output of vitamin C during the febrile stages of these diseases is certainly not reduced.

Godber's assumption that the convalescents can be regarded dietetically as normal people really begs the question: for in conditions where the tissues are heavily depleted of vitamin C - as, for example, in Harris's experiments mentioned on page 6 - it takes considerably more than a fortnight of adequate diet to restore the lost reserves.

Moreover, in the light of the knowledge of vitamin C requirements and standards that has accumulated in the four years subsequent to Godber's experiments, the first point that strikes one is that, for " normal people" receiving a diet fairly generous in vitamin C, practically every convalescent had a remarkably low output of the vitamin.

At the time when these experiments were carried out Harris's original standard of adequacy - a concentration above .01 to .02 mg./cc. of urine, or a total adult output of above 13 to 27 mg. - was still accepted. Godber realised that this standard, being founded on a " reputed minimal optimal dose" which itself was based on the quite valueless cuff test, had no valid basis, but, unfortunately, he thought it was too high, and therefore disregarded the fact that half of his convalescents were below even Harris's minimum figure. As has been shown, Harris's original standard was, in point of fact, dangerously low: a level of .01 mg. of vitamin C per cc. of urine (or a total output of 13 mg.) is not even incompatible with the development of scurvy. Yet, out of 73 convalescents examined, 35 has a concentration lower than .01 mg./cc.; and,of 48 convalescents whose total excretion was measured, that of 25 fell below the 13 mg. line.

If we accept a concentration of .015 mg./cc or an adult excretion of 19.5 mg. as the lowest possible standard of normality (see page 20), - a standard far below what we would expect in normal people who were fed according to the generous diet of an L. C. C. hospital -, then, out of 73 convalescents, only 14 reached a normal concentration (and of these only 4 were above normal). Concentration, however, is an uncertain guide, since it may vary according to the amount of urine passed. On the safer ground of total excretion, out of 48 convalescents whose output was calculated, only 4 reached a standard of normality. (These 4, incidentally, consisted of two mild cases of diphtheria and 2 of scarlet fever.)

When we find that, after at least a fortnight's convalescence during which they received a diet rich in vitamin/

vitamin C, only 4 mild cases attained anything like a normal excretion, as compared with 44 whose output was grossly subnormal, we can only conclude that the depletion of the ascorbic acid reserves during the acute stage must have been very considerable.

No useful purpose can be served by comparing the excretion during the acute stage with the markedly sub--normal excretion of convalescence.

Taken by themselves, the concentrations during the acute stage do not seem to be much diminished, except in Scarlet Fever (where half of the patients had a concent--ration below the lower limit of normality), and in the Puerperal group (where only one patient attained a normal level); but the possibility of febrile oliguria makes the concentration - at no time a really reliable guide - a wholly unreliable index. Adopting the safer criterion of standardised excretion, we find that the excretion was sub--normal in 66.7 % of the acute cases of scarlatina, 55 % of the acute cases of diphtheria, 100 % of the puerperal cases, and 60 % of tonsillitis cases. These figures, however, mean little, as we do not know the previous nutrit--ional state of the patients.

In support of his contention that infection does not lower the excretion of vitamin C, Godber points out that there is no correlation between the excretion and either the severity of the case or the degree of pyrexia; but, in respect of the excretion at the start of the illness, the possibility of any such correlation is ruled out by the presence of another variable, namely, the previous levels of ascorbic acid in the tissues of the patients. It is only in patients who had received similar diets prior to the onset of the infection that we could expect any such correlation. As for the excretion in convalescence, in view of the many possible fallacies of the dye test, the numbers are too small for us to expect to find a high degree of correlation; yet it is interesting that, out of 10 scarlet fever patients with an initial temperature of 100° or more, 7 had a convalescent concentration below the average for the convalescent scarlet group; and that, out of 8 cases of severe diphtheria, 5 had a convalescent concentration below the average for that disease. Even more interesting is an analysis of the 15 diphtheria cases of which the total excretion was ascertained during convalescence:

	No. of cases with total excretion		
	Under 13 mg.	13 - 19.5mg.	Over 19.5mg.
8 severe or moderate cases	5	3	0
7 mild cases	3	2	2

Incidentally/

Incidentally, Godber's cases of puerperal pyrexia suggest a relationship between the duration of the pyrexia and the total daily excretion of ascorbic acid:

Duration of Pyrexia.	Vitamin C excretion.	Vitamin C output two days later.
88 days	10.2 mg.	---
48 "	11.1 mg.	---
19 "	13.6 mg.	11.7 mg.
16 "	12.4 mg.	13.0 mg.
8 "	---	---
3 "	16.9 mg.	13.5 mg.

Godber's error in assuming that his convalescents, after "at least a fortnight" of full hospital diet, could be regarded, in respect of ascorbic acid nutrition, as normal people - a mistake easy enough to make in 1938 when knowledge of the normal standards for vitamin C was most incomplete - invalidates his conclusions but does not detract from the sterling value of his work: although he employed only a single test of very limited accuracy, his careful study of the excretion of ascorbic acid in infect--ious diseases is recorded with such precision and detail as to make it possible for subsequent workers, utilising information unknown in 1938, to draw valid conclusions from his experiments. - In particular, the fact that, after a full fortnight on a generous diet, 91.7 % of his convalescents had a grossly subnormal excretion of the vitamin affords additional evidence of the extent to which the bodily reserves become depleted in infectious diseases.

(g) Conclusions regarding application to infections of standards for vitamin C.

All the evidence indicates that infectious disease lowers the ascorbic acid content of the tissues. While this at once suggests that, in order to restore the tissue concentrations to normal, the diet of convalescents should be rich in vitamin C, we have still to consider whether the depletion is so drastic as to warrant administration of the vitamin during the acute stage.

Yavorsky and others (152), Ingalls (153), Marinesco et alii (154), Sweeney and his co-workers (125), and others have investigated the amount of ascorbic acid in various organs and fluids. On the basis of such work the vitamin C content of the adult body may be assessed at about 1,000 mg. in an average person and about 3,000 mg. in a saturated person. Of the normal 1,000 mg. of vitamin C, some 200 - 250 mg. are stored in the liver, about 20 - 30 mg. are in the blood, and considerable amounts are present in the adrenals, brain, kidney, pancreas, spleen, heart and lung.

Now/

Now Falke, as already mentioned on page 24, found that, to maintain the blood ascorbic acid at a constant level during a febrile illness, an extra 100 mg. were required daily. If his figures are accepted as accurate, it follows that, where pyrexia lasts for not more than three days, failure to supply extra vitamin C will result in the tissues becoming depleted only to the extent of less than one third of their normal total content.

The provision of additional ascorbic acid during prolonged febrile illnesses is an obvious necessity: as a recent leading article in the 'British Medical Journal' (126) put it, speaking of pulmonary tuberculosis, -

> " Without supplementary administration of vitamin C, the store (of the vitamin) is dissipated in advanced tuberculosis before death, sometimes to a negligible quantity."

For fevers of short duration, however, such as varicella and the mild scarlatina of recent years, the suggestion that " the generally accepted amount of 50 to 60 mg. daily" should be increased to about 200 mg. (126) seems to lose force. Unless it can be shown that administration of vitamin C during the febrile period (when the amount of waste through non-absorption is likely to be considerable) favourably influences the course of the disease, it may well be maintained that, for fevers of short duration, no harm will be done by allowing the body to become tempor--arily depleted of one third of its vitamin C, so long as the diet in convalescence (when absorption is easier) contains not merely sufficient ascorbic acid for normal maintenance but a surplus to enable the bodily stores to be built up again. For transient pyrexias, then, a convalescent diet containing ample fresh fruit and green vegetables may well suffice: administration of synthetic vitamin C during the acute stage cannot be regarded as necessary unless it can be shown that such administration has a beneficial effect during the actual period of illness.

(3) EFFECTS OF VITAMIN C " IN VITRO ", AND EXPERIMENTS ON LABORATORY ANIMALS.

Although logically this section should be divided into two sub-sections, dealing with " test-tubes " and " guinea-pigs " respectively, it is felt that a clearer picture will be obtained by taking various organisms separately.

(A) H. Pertussis.

Otani (155 & 156) claimed that, when cultures of H. Pertussis were grown on solid media, an addition of 2.5 mg. /

mg. of vitamin C to every cc. of medium had a definite retarding effect on the growth of the organisms; and when the amount of ascorbic acid was increased, the inhibitory effect became more marked, until finally death of the organisms occurred. Similar results were reported almost simultaneously by Grootton and Bezsonoff (157). It is to be noted, however, that the concentration required to produce this retarding effect is tremendously high, about a hundred times as high as we could hope to achieve in the circulating human blood, and that the inhibitory action might conceivably be due merely to alteration in the pH of the medium.

Otani also found that, when the quantity of vitamin C added was 1.2 - 1.8 mg./ cc. of medium, normal growth occurred, but the organisms were of diminished virulence: when injected into rabbits they caused only a leucocytosis, instead of the expected leucocytosis with relative lymph--ocytosis. He showed, too, that, if ascorbic acid were added to the toxin of H. Pertussis, the effect of the latter when injected intradermally into rabbits or guinea--pigs was markedly reduced; and that, if the untreated toxin were injected into animals which had previously been dosed with ascorbic acid, the effect was again diminished.

(B) C. Diphtheriae.

As early as 1935, Jungeblutt and Zwemer (158) claimed that vitamin C had some power of inactivating diphtheria toxin, and that animals treated with the vitamin reacted less severely to subsequent intradermal injections of the toxin. Following up this line of investigation, Kligher and his co-workers (159) found that the addition of vitamin C to cultures of C. Diphtheriae reduced their toxigenicity, as estimated by intradermal tests in the same animals. They also contended that preformed diphtheria toxin was, *in vitro*, partially inactivated by the addition of ascorbic acid. Attempting to refute this last claim, Torrance (160) suggested that the rapid disappearance of the vitamin in incubated toxic filtrates is due to the toxin rapidly inactivating ascorbic acid by a reversible oxidation.

Investigating the effects of a lethal dose of toxin, Harde and Benjamin (161) ascertained that, in guinea-pigs dying from the effects of an injection of diphtheria toxin, the ascorbic acid content of the suprarenals and liver was markedly subnormal; and these organs (together with the brain and kidneys) are the very ones in which the ascorbic acid content falls sharply when guineapigs are fed on a hypovitaminous diet, - as has been shown by Yavorsky and others (152), Malmberg and Euler (162), and Wortis, Wortis, and Marsh (163).

This dramatic fall in the ascorbic acid content of the /

the suprarenals and liver after a fatal dose of diphtheria toxin was confirmed by Kligher et alii (159), by Careddu and Wilzer (164), and by Harris, Passmore and Pagel (165). The last named workers found that, although the fall was more striking after diphtheria toxin, a similar fall could be produced by injecting the toxins of certain members of the Salmonella and Pasteurella groups or by the injection of M. Tuberculosis; and Harde and Benjamin - following up their original experiment - discovered that the liver and suprarenals were largely depleted of vitamin C after the injection of the toxin of diphtheria, tetanus, dysentery, or mouse typhoid.

Torrance (166) corroborated that a lethal dose of diphtheria toxin reduced the ascorbic acid content of the suprarenals almost to nil. He also found that, curiously enough, the injection of half of a minimum lethal dose caused a rise in the suprarenal content within forty-eight hours, while one third of a minimum lethal dose caused a rise within twenty-four hours.

Flori (167) reported that administration of vitamin C benefited guinea-pigs suffering from diphtheria, and he suggested that the vitamin enhanced the action of diphtheria antitoxin. More recently, Luck and Hall (168) have confirmed Jungeblutt's original suggestion that guinea-pigs treated with vitamin C withstand diphtheria toxin far more satisfactorily than untreated controls.

Other workers (169 & 170) have used ascorbic acid along with adrenal cortex hormone, and have claimed that diphtheritic toxaemia is less severe in rabbits so treated than in control animals.

Finally, Szent-Gyorgyi's experiments on guinea-pigs " Show that whereas 1 mg. daily prevents the onset of obvious signs of scurvy, about 2 mg. is needed for normal tooth development, and 3 or more to protect the animal from injury by injection of diphtheria toxin." (171).

(C) M. Tuberculosis.

McConkey and Smith (172) kept 37 guinea-pigs on a diet containing no vitamin C and 25 guinea-pigs on a diet rich in the vitamin. All the animals were then fed with tuberculous sputum. Only two of the 25 guinea-pigs which had received ample ascorbic acid developed tuberculous enteritis, as compared with twenty-six cases in the 37 hypovitaminous animals, - a difference which is, of course, statistically significant.

That vitamin C afforded some protection against tuberculosis was denied by Heise and Martin (173): they induced /

induced a hypervitaminosis C in 10 out of 15 guinea-pigs, and subsequently infected all the animals with tuberculosis. However, even if the inadequacy of the numbers tested be disregarded, this experiment proves little: a dose of M. Tuberculosis sufficiently potent to infect every single guinea-pig is obviously no criterion by which to decide whether the state of vitamin C nutrition has any bearing on the animals' powers of resistance to infection.

Similarly, the finding that prolonged medication with vitamin C does not protect guinea-pigs against a lethal injection of tuberculin (174) is not evidence for or against the claim that animals treated with the vitamin have a greater power of resistance than untreated controls. Indeed, the very workers who first showed that ascorbic acid does not confer immunity against a lethal dose of tuberculin have recently produced evidence (175) to prove that daily injections of vitamin C benefit tuberculous guinea-pigs.

Birkhaug's conclusions are in substantial agreement both with these claims regarding the therapeutic effect of ascorbic acid and with McConkey's findings regarding its prophylactic effect. Birkhaug (176) reported that hyper-vitaminosis C caused a significant increase in the body weight of guinea-pigs and a reduction in the frequency of tuberculous lesions; and that daily oral administration of the vitamin significantly inhibited the tuberculin reaction in tuberculous guinea-pigs, while the degree of inhibition was correlated with the level of urinary excretion and with the suprarenal content of vitamin C.

These reports are reinforced by the observations of Osborn and Gear (177), who have pointed out that those mammalian species which can synthesize vitamin C are the ones most resistant to tuberculosis.

(D) Coccaceae.

Mayer (178) showed that vitamin C inhibits the growth of pneumococci and streptococci. Earlier workers (179 & 180) had commented on the fact that, when an animal is deprived of ascorbic acid for a long time, instead of developing scurvy, it usually develops pneumonia.

Rhinehart (181) asserted that, if guinea-pigs were deprived of vitamin C and then infected with haemolytic streptococci, the resultant lesions closely resembled those found in rheumatic fever in man; but subsequent workers have pointed out that Rhinehart's lesions were simply those found in guinea-pigs deprived of vitamin C: they were not lesions due to streptococcal infection.

Busing (182) found that daily intramuscular or intravenous /

intravenous injections of ascorbic acid into rabbits or rats infected with pneumococci increased their survival time, as compared with that of controls which received no extra ascorbic acid; and Locke and his co-workers (183) claimed that the administration of vitamin C improved the resistance of rabbits to Type I Pneumococcus, and also enhanced the action of sulphanilamide treatment.

For pneumococcal lesions in experimental animals, as for tuberculous and diphtheritic conditions, there is, therefore, a fair amount of evidence in support of the view that ascorbic acid is beneficial.

(E) Other Organisms.

Patocka and Ilavsky (184) have shown that, *in vitro*, ascorbic acid promotes the growth of strict anaerobes, a result, presumably, of its reducing property; but, since this enquiry is not concerned either with the genus Clostridium or with the common cold - the virus of which is said (185) to be a strict anaerobe - the effect of the vitamin on anaerobes is hardly relevant.

Jungeblutt (186) has claimed that, within certain narrow limits of concentration, ascorbic acid inactivates the virus of poliomyelitis. He has also reported (187 & 188) that treatment with the vitamin confers some measure of protection on monkeys incubating poliomyelitis after experimental infection. However, Sabin (189) has been unable to find any evidence of even partial protection in similar circumstances.

(F) Anaphylactic Shock.

In 1939 Diehl (190) investigated the effects of different states of vitamin C nutrition on anaphylaxis. He found that, in guinea-pigs suffering from a mild hypo--vitaminosis C, anaphylactic shock was more severe than in normal animals, and more frequently ended fatally; on the other hand, in guinea-pigs suffering from a severe degree of hypovitaminosis (or from avitaminosis), an anaphylactic reaction was hard to produce, and, when produced, was relatively mild. Diehl tried to explain his findings by postulating that in mild hypovitaminosis the reticulo-endothelial system produces a small amount of antibodies, whereas in avitaminosis it produces practically none.

Subsequently, a Japanese worker (191) discovered that, in guinea-pigs sensitised to horse serum, he could prevent or minimise anaphylaxis by giving an injection of ascorbic acid immediately before the second dose of horse serum.

(4) THERAPEUTIC ADMINISTRATION OF VITAMIN C IN INFECTIONS.

(A) Tuberculosis.

The evidence in regard to tuberculosis is abundant but conflicting, although the great majority of published reports indicate that the therapeutic administration of ascorbic acid is not devoid of beneficial results.

In America, Radford and his co-workers (192) divided 111 cases of advanced pulmonary tuberculosis into three groups: patients in Group C received 250 mg. of vitamin C daily, members of Group O were given 500 cc. of orange juice, and Group Co was a control group receiving no dietetic supplement. At the end of a three months course of treatment, Group C showed an improvement over the control group in respect of haemoglobin, erythrocyte count, monocyte/lymphocyte ratio, and neutrophil/lymphocyte ratio, while Group O was intermediate between the other groups; but repetition of these tests some three months later showed that the differences had been transient, and clinical examination failed to reveal any significant difference between the three groups. In Austria, Weber (193) claimed that administration of ascorbic acid caused a definite regression of infiltration. In Germany, Pilz (194) and Hasselbach (195 & 196) noted that treatment of active pulmonary tuberculosis with vitamin C was followed by definite clinical improvement, with better appetite, increase in weight, and reduction of night sweats. In Italy, Capelli (197) found that daily intravenous injection of vitamin C for ten doses was useful for certain cases of haemoptysis due to pulmonary tuberculosis. Writing in the "British Journal of Tuberculosis", Heise and others (198) maintained that five daily intravenous injections of the vitamin produced a fall in the blood sedimentation rate. Improvement in the blood sedimentation rate and increase in the body weight were recorded by Albrecht (199) as beneficial results of the administration of ascorbic acid. Similar claims for slight degrees of improvement have been made by Petter (200), Hurford (138), Bakhsh and Rabbani (201), etc.

Incidentally, it should be noted that, if - as Schneider and Widman (202) have reported - high dosage with vitamin C lowers the blood sedimentation rate of normal individuals, the fact that administration of the vitamin reduces the high blood sedimentation rate of pulmonary tuberculosis does not, *per se*, necessarily indicate any beneficial action as regards the actual disease.

Again, Sweany (125) found that ascorbic acid was very useful in advanced tuberculosis, decreasing sputum, improving general /

general condition, and prolonging life; and Cruickshank and Feaver (203) postulated the presence of a " Vitamin C hunger " in active tuberculosis.

On the other hand, Dagulf (204), after a careful and comprehensive study of past work and of his own experiments, concluded that - although the vitamin C content of blood and urine is definitely reduced in tuberculosis - there is no proof that ascorbic acid therapy is beneficial. This conclusion is supported by the work of Erwin, Wright and Doherty (141), who endeavoured to treat seven acute cases with marked pyrexia and seventeen " bad chronics " with 100 - 200 mg. of vitamin C daily for six months: of the twenty-four cases so treated, eleven died during the investigation, and of the remaining thirteen - all of whom remained sputum positive - the results might be summarised as follows:-

	Improved.	Unchanged.	Worse.
X-ray appearance	1	0	12
Pyrexia	2	7	4
Pulse	2	7	4
Amount of sputum	0	5	8
Weight	3	1	9.

As for complications,-

> " Several of the patients were suffering from laryngitis or enteritis, but in none of the survivors was any improvement in these complications noted ";

two patients developed haemoptysis while under treatment, and one developed meningitis. Admittedly the study of Erwin and his co-workers suffers from certain defects: the number of cases investigated is small, all the cases were very advanced, and the absence of controls is most unfortunate. Yet these defects hardly seem sufficient to warrant the complete rejection of the testimony of the three Liverpool workers.

The investigators who maintain that ascorbic acid is not beneficial - Erwin and his colleagues, Dagulf, and one or two writers quoted by Dagulf - may form only a minority, but they are a minority too strong to be ignored. While it is indisputable that in tuberculosis the body becomes depleted of its store of vitamin C, and while measures to remedy that depletion seem entirely reasonable, the question of whether the administration of ascorbic acid does anything to reduce the severity of the disease must be regarded as still open.

(B) Rheumatic Fever.

In view of the low level of vitamin C in urine and in plasma in cases of rheumatic fever, a number of workers have endeavoured to ascertain whether administration of the /

the vitamin would have any effect on the course of the disease. Schultz (6) divided 56 advanced rheumatic cases into two equal groups: members of Group A received 100 mg. of vitamin C daily, while the patients in Group B were given 100 mg. of lactose; and there were more relapses in Group A than in Group B. He then tested the effect of intravenous and oral ascorbic acid on 44 cases of acute rheumatic fever, and found no clinical improvement in patients so treated.

With these conclusions Faulkner (205), Sendroy and Schultz (131), and others have agreed: the clinical manifestations of acute rheumatic fever are not demonstrably affected by oral or intravenous administration of vitamin C over periods of several months, or by large doses of orange juice.

(C) Whooping Cough.

As contrasted with the doubtful results obtained in pulmonary tuberculosis and the negative results in acute rheumatism, the findings in whooping cough are positive: administration of vitamin C to patients suffering from whooping cough has been uniformly recognised as beneficial by the few workers who have published reports of their investigations of this line of treatment.

Otani (155) employed parenteral vitamin C as the only treatment in 81 cases of whooping cough, including some with associated bronchitis or pneumonia. For mild cases he gave intravenous or intramuscular injections of 50 - 100 mg. daily for 5 - 12 days, and for severe cases rather larger injections for a similar period. In 66 cases (or 81.5 %) the results were good, the spasmodic cough improved greatly after one to two weeks, and the blood picture approximated to normal within ten days. While it was with uncomplicated cases that ascorbic acid treatment was most successful, three severe cases " in which a fatal issue was threatened " recovered under this treatment. In the 15 cases (or 18.5 %) in which the ascorbic acid therapy was unsuccessful there were usually complications, such as asthma, tuberculosis, or simult--aneous measles. On the basis of these results Otani concluded that,

> " Treatment with vitamin C may therefore be regarded as a specific procedure. In contrast with other methods of therapy, this treatment shows no side effects, even when unnecessarily large doses of vitamin C are given. It possesses the advantage, when used for pertussis in infants, of giving good results at an age when, by reason of the inadequate production of antibodies, success with specific vaccines is difficult to obtain."

Ormerod /

Ormerod, UnKauf, and White (146) treated 17 cases of pertussis with oral vitamin C, the dosage given being - irrespective of age - 350 mg. on the first day, 250 mg. on the second and third days, 200 mg. on the fourth and fifth days, 150 mg. on the sixth and seventh days, 125 mg. on the eighth and ninth days, and then 100 mg. daily till all symptoms vanished. They found that treatment with vitamin C caused changes in the following order:

" (1) Marked reduction or complete arrest of vomiting;
(2) Reduction or disappearance of night cough;
(3) Reduction in number or intensity of whoops;
(4) Reduction in number or intensity of day coughs."

They concluded that ascorbic acid therapy reduced the duration of the disease from a matter of weeks to a matter of days.

These experiments were perhaps open to criticism on the ground of absence of controls. This criticism is not, however, valid in respect of Gairdner's investigation (206). Gairdner studied 41 cases which fulfilled one or more of the following conditions: (a) H. Pertussis recovered from cough plate; (b) Typical paroxysmal cough witnessed; and (c) Suggestive history accompanied by either sublingual ulcer or marked lymphocytosis. Of these cases, 21 received vitamin C, the dosage employed being 200 mg. daily for the first week, 150 mg. daily for the second week, and 100 mg. for the third week. The other 20 cases received cod liver oil, bromide and belladonna. Although the vitamin C was not always administered at the start of the disease, Gairdner found that there was a significant decrease in the total duration of illness in the group treated with ascorbic acid: the average duration in the treated group was 35 days, as compared with 41 days in the control group.

(D) Pneumonia.

As an introduction to the report of their experiments Gander and Niederberger (63) gave an interesting summary of the evidence that led them to enquire whether ascorbic acid therapy might be beneficial in pneumonia. Their main points are:

1. The time-honoured administration of fruit and fruit juices in febrile conditions, including pneumonia;
2. The surprisingly high degree of vitamin C deficiency commonly found after pneumonia;
3. The frequent occurrence of pneumonia in animals fed on scurvy-producing diets;
4. The high correlation between the seasonal mortality from pneumonia and the seasonal frequency of vitamin C deficiency;

and 5. The increased mortality from pneumonia in the elderly, in whom vitamin C deficiency is generally most marked.

They /

They treated 15 cases of pneumonia with vitamin C orally or intramuscularly. Usually the administration of ascorbic acid was additional to the ordinary pneumonia therapy, but in one case oral treatment with vitamin C was successfully applied without any other medicaments. The findings in these 15 cases suggest that ascorbic acid, particularly if given early, favourably influences the course of the disease: pain subsides, thus permitting of a reduction in the exhibition of narcotics, and

> " The general condition of the patient is always benefited in a striking manner, and convalescence runs a better and more rapid course than in pneumonias which are not treated with vitamin C. An impaired percussion note, bronchial breathing and adventitious sounds certainly persist for a while, since the healing process cannot immediately follow on the resolution of the diseased tissues. No cases of failure have yet been recorded, although the condition of some of the patients treated was very serious."

Gander and Niederberger varied their dosage according to the amount of vitamin C deficiency as estimated by urine tests. On an average, however, their recommended dosage is in the neighbourhood of 2,000 mg. in one day, - for cases with gastro-intestinal disturbances about four injections of 500 mg., and for cases without alimentary upset, one intramuscular injection of 500 mg. followed by 1,200 mg. orally during the next few hours.

They concluded that,

> " Correction of the state of vitamin C deficiency on the first day of the attack of pneumonia brings about such strikingly favourable results as to warrant the inference that vitamin C represents a valuable addition to the therapy of pneumonia. "

As a corrective to these enthusiastic claims it must be pointed out that Gander and Niederberger have recorded details of only four of their fifteen cases, and that, even for these four, the information provided is inadequate.

Hochwald (207) treated 13 cases of croupous pneumonia solely with intravenous injections of ascorbic acid. He found that

> " The improvement in general condition (prostration, dyspnoea) is particularly striking; it becomes apparent after the very first injections, and is quite marked even while the pyrexia persists. The more rapid return of temperature to normal is another feature; it is accompanied, or often preceded, by a sudden decrease in the leucocyte count /

" count, and morphological improvement of the blood picture. The clinically, or more often radiologically, demonstrable infiltration of the lung parenchyma, which usually persists for several weeks after the temperature has returned to normal, retrogressed very early in our cases. "

Hochwald's dosage was 500 mg. every two hours until the temperature fell.

Vogl (208) treated lobar pneumonia and broncho-pneumonia with ascorbic acid, giving 200 - 500 mg. twice daily for several days. In lobar pneumonia he found that remarkable changes were observed in the temperature chart: within 24 hours the morning temperature was normal; and during each day the temperature rose to a lower maximum than on the previous day, until on the fourth or fifth day it remained normal. Also, cyanosis, dyspnoea, and restlessness were less marked than in other cases. The physical signs, however, did not disappear any more quickly than in untreated cases.

In broncho-pneumonia, too, the symptoms were less troublesome, and the temperature behaved as in lobar pneumonia, except that it took longer to reach normal.

In all, Vogl had two deaths in his 37 cases of pneumonia treated with vitamin C.

Vogl also claimed that vitamin C was a useful prophylactic against the development of post-operative pneumonia, a claim that has recently been refuted by Lund and Crandon (102).

(E) Diphtheria.

The evidence relating to diphtheria is confused and contradictory, and no worker has yet produced a series of treated cases and controls of sufficient magnitude to decide the case one way or the other.

As early as 1935, Bamberger and Wendt (147) attempted to treat malignant diphtheria with vitamin C. In view of the well known post-mortem finding of pathological changes in the suprarenals in fatal cases of diphtheria, and in view of the equally well known fact that neither large doses of antitoxin nor circulatory stimulants will prevent circulatory failure, these workers had previously tried to treat circulatory failure with extract of suprarenal cortex, but without success. They now continued to administer suprarenal extract, but - because the normal suprarenal is especially rich in vitamin C - they also gave ascorbic acid, in doses of about 500 mg. per day for two days, usually orally.

Applying /

Applying this treatment - suprarenal extract and vitamin C to eight clinically hopeless cases, they were able to record five recoveries, while in the three failures " death was postponed four to five days."

Following up this line of treatment, Bamberger and Zell (209) reported that, out of 41 cases of very toxic diphtheria treated with suprarenal extract and ascorbic acid, 22 had survived, - a result which Bamberger and Zell regarded as satisfactory, but which seems meaningless in the absence of any statement of the mortality rate for equally toxic cases when given no suprarenal extract or ascorbic acid.

Mautner (169) and Dieckhoff (170) concluded separately that, although vitamin C and suprarenal extract definitely benefited experimental animals, clinical trials on human beings were not very encouraging. Even more disappointing were the results of Engelhard (210) who used the alternate case method: of 28 patients treated with vitamin C and cortical extract, seven died; in 27 controls there were eight fatalities.

On the other hand, Otto (211) revived the claim that both ascorbic acid and suprarenal extract appeared to exercise a favourable influence on the course of malignant diphtheria.

(F) Other infections.

Heaslip (212) found that a low state of vitamin C nutrition predisposed to infection by the virus of poliomyelitis and also increased the severity of attack. Korbsch (213) claimed that ascorbic acid in large doses gave excellent results in the treatment of head colds and acute rhinitis. Dainow (214) treated a few cases of herpes zoster by daily injections of 100 mg. of vitamin C, and found that, under this treatment, the lesions rapidly disappeared and the pain subsided.

In an investigation involving 300 cases Glazebrook and Thomson (95) showed that ascorbic acid reduced the duration of tonsillitis. Baer (145) successfully treated 36 cases of pharyngitis with the vitamin. Kellett (216) advised the use of vitamin C - in addition to neoarsphenamine - in Vincent's angina. Villela (216) found that in skin leprosy vitamin C had little real value, apart from possibly increasing slightly the tolerance to chaulmoogra preparations. Gerdjikoff (217) studied twelve cases of malaria treated with ascorbic acid in addition to ordinary antimalarial drugs: he concluded that ascorbic acid, given in addition to specific therapy, was very beneficial in malaria.

Najib-Farah /

Najib-Farah (218) treated enteric fever with intra--venous injections of vitamin C and extract of suprarenal cortex, and reported that this treatment gave immediate beneficial results. And Cammarella (219) found that in dysentery administration of ascorbic acid was valueless.

With the exception of the work of Glazebrook and Thomson, the experiments mentioned in the previous three paragraphs were all conducted on a limited scale, and the various results are suggestive, rather than conclusive.

(G) Experimentally induced fever.

Haas (220) analysed the effect of ascorbic acid on experimentally produced pyrexias, and showed that, while the vitamin prevents a rise of temperature in those cases where the pyrexia is due to drugs which affect the peripheral temperature regulation (e.g. dinitrophenol and thyroxin), it does not prevent the rise of temperature in cases where the pyrexia is caused by drugs which act through the central nervous system (e.g. benzidrine and pyrifer).

(5) OTHER POINTS OF INTEREST.

If ascorbic acid therapy is alleged to be beneficial in certain zymotic diseases, there immediately arises the question of how ascorbic acid can produce a beneficial effect. In this section it is proposed to touch briefly on certain physiological points.

(A) The erythrocyte in relation to infectious diseases and to vitamin C.

It is a well known fact that a hypochromic, microcytic anaemia occurs in typhoid, diphtheria, and " to some degree in most specific fevers " (221). Now, the administration of vitamin C not only cures the very similar anaemia found in scurvy (222 & 223), but also - according to a standard text-book on diseases of the blood (224) - causes a reticulocyte response in patients suffering from infections.

Also, it is generally admitted that vitamin C is essential to normal erythropoiesis (225).

(B) The leucocyte.

A falling leucocyte count has long been recognised as a grave prognostic indication in such diseases as lobar pneumonia and cerebrospinal meningitis; and the introduction of sulphapyridine therapy has brought agranulocytosis into prominence /

prominence. Hence, any drug which will prevent or cure leucopenia (and, in particular, polymorphonuclear leuco--penia) might find a place in the therapy of infectious diseases.

That vitamin C is of value in leucopenia was first demonstrated by Schnetz (226), who found that, although administration of the vitamin produces no leucocytosis in normal persons, such administration causes a definite rise in the leucocyte count in cases of leucopenia. Again, Kalk (227) has reported six cases of agranulocytosis successfully treated by daily injections of ascorbic acid. And Carrie (228) has claimed that vitamin C given during a course of X-ray therapy prevents an irritation leucopenia.

(C) Antibodies, etc.

The relationship of vitamin C to immunity processes is far from clear. It has been shown that there is a correlation between the concentration of ascorbic acid in the blood and complement in the serum (229); it has been proved that vitamin C increases the phagocytic properties of blood (178); and it has been claimed that the production of specific antibodies can be stimulated by intravenous injection of the vitamin (229).

(D) Glycogenolysis.

If the much favoured glucose drip has any value in severe infections, then a drug which influences the level of glucose in the blood must be important. It has been shown (230) that glycogen formation in the liver of hypo--vitaminous rabbits is greatly stimulated by vitamin C; and in human beings it has been found (231) that the rise in blood sugar of a patient who took a hundred grammes of glucose by mouth was greater after high dosage with vitamins B and C than after no vitamin therapy.

(E) Detoxicating function of vitamin C.

The detoxicating action of ascorbic acid is well established: for example, Holmes and others (232) have demonstrated that it is beneficial in lead poisoning; Bise (233) and Lees (234) have found it useful in arsenical dermatitis; and Buckley and Sande have both recommended it for the treatment of cutaneous reactions following gold therapy.

While these findings are of significance in regard to syphilis, tuberculosis, and rheumatism, the action of ascorbic acid in preventing or reducing the toxic effects of the sulphonamide compounds is important in regard to such infections as cerebrospinal meningitis, tonsillitis and pneumonia. Not only does vitamin C have a valuable action/

action in preventing agranulocytosis due to these drugs, but - according to Dainow (235) - intravenous injections of the vitamin prevent or diminish all the other toxic effects of the sulphonamide compounds.

There are other points in which the physiological actions of ascorbic acid suggest that it might have some value in infectious conditions - e.g. administration of the vitamin is followed by an increase in the lipase content of the blood (236), and a similar rise has been noted in the lipase content of the blood of tuberculous patients when they are progressing towards cure (237); however, the points already specified should be sufficient to demonstrate that there is an adequate physiological basis for clinical trials of ascorbic acid in infectious diseases.

CONCLUSIONS FROM PART 1.

(1). Although early workers used inaccurate methods for the estimation of vitamin C, and suggested different (and more or less arbitrary) standards of adequacy and normality, accurate methods of assay are now in existence, and - within fairly narrow limits - generally accepted standards have been devised for human requirements of the vitamin and for the vitamin C content of various body fluids and tissues. Hence, it should be possible to determine whether, in any particular disease, the bodily stores of the vitamin are reduced.

(2). A survey of the published work shows that the bodily reserves of ascorbic acid are considerably reduced in Whooping Cough, Pneumonia, Diphtheria, Scarlet Fever, Rheumatic Fever, Pharyngitis, and Tuberculosis. For other infections evidence is lacking.

(3). Since the tissues are partially depleted of vitamin C during the acute stage of these seven diseases, it follows logically that this depletion should be made good either by the administration of large doses of the vitamin during the acute stage or by the provision during convalescence of a diet rich in ascorbic acid; but (except, perhaps, in tuberculosis, where the acute stage is of long duration) it does not necessarily follow that the former of these alternatives is preferable to the latter.

(4). Experiments on laboratory animals suggest that the administration of vitamin C may be beneficial in Whooping Cough, Pneumonia, Diphtheria and Tuberculosis. There is no evidence regarding other infections.

(5) A consideration of some of the physiological functions of /

of vitamin C makes it seem quite probable that administ--ration of the vitamin will be useful in some of the more serious infections.

(6). For most infectious diseases the efficacy of vitamin C therapy has still to be proved or disproved by clinical trial: the position is best summarised in the words of Harries and Mitman (Clinical Practice in Infectious Diseases, 1940, p.89), - " There is a loss of vitamin C in fevers, and it has been suggested that vitamins B and C have a therapeutic value, but this is not yet proved." In many infections (e.g. Cerebrospinal Fever, Erysipelas, Scarlet Fever, Influenza, Measles, Chicken-Pox, Small-Pox, Mumps, Glandular Fever, Undulant Fever, Typhus, etc.) ascorbic acid does not appear to have been used; in other infections (Enteric, Coryza, Tonsillitis, Pharyngitis, and Malaria) isolated workers have reported favourably on the effect of the vitamin, but these reports still await confirmation or refutation. In only five infectious diseases has ascorbic acid been given a reasonable therapeutic trial, - Rheumatic Fever (if the term "infectious" can be stretched to cover this condition), where vitamin C has proved valueless; Whooping Cough and Pneumonia, where it appears to be beneficial; and Diphtheria and Tuberculosis, where the published results are conflicting.

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PART 2 - AN INVESTIGATION OF THE EFFECTS OF ASCORBIC ACID THERAPY IN CERTAIN INFECTIONS.

In 1939 and 1940, for a period of about a year, the writer attempted to investigate the effects of ascorbic acid therapy as applied to infectious patients, practic--ally all of whom were in his own clinical charge, in Edinburgh City Hospital. The patients studied included 108 cases of influenza, 148 cases of cerebrospinal fever, 114 cases of diphtheria, 32 cases of streptococcal tonsillitis, 40 cases of scarlet fever, and 14 cases of post-influenzal pneumonia. In each disease the patients were divided into two comparable groups, which received the same diet and treatment, except that one group was given synthetic vitamin C. Subsequently, in 1941 and the early months of 1942, a few confirmatory experiments were carried out on patients under the writer's clinical charge at Kendray Isolation Hospital, Barnsley. The patients studied in these confirmatory experiments included 36 cases of diphtheria and 16 of cerebrospinal meningitis.

The means taken to ensure that the groups were really comparable, and also to eliminate the effects of any unconscious personal bias on the part of the writer, are discussed in the reports on the separate diseases.

(1) INFLUENZA.

The occurrence of an outbreak of influenza in two ships of the Navy while in port, and the consequent admission to hospital of 108 seamen, provided a golden opportunity for obtaining comparable groups. The patients, who were similar in sex, occupation, previous diet, previous general health (the high standard required for the Senior Service), etc., were transferred to hospital 24 - 48 hours after first reporting sick. They all suffered from a mild influenzal syndrome, - pyrexia, malaise, pharyngitis, some tracheitis, and vague pains; and they were obviously infected under very similar conditions.

All patients were put to bed, and treated with Dover's powder and aspirin (aā gr.X, bis), throat gargles, and purgatives; all received identical diet; and every second case was given ascorbic acid orally, - 50 mg. four times daily for two days. No additional treatment was given except in cases which developed complications.

Two criteria have been used in comparing the two groups/

groups, - (a) the incidence of complications and relapses, and (b) the duration of illness from the date of reporting sick to the date of discharge from hospital. No question of personal bias arises over the discharging of the cases, because the discharges were arranged not by the writer but by a colleague (the senior assistant medical officer at the hospital) who was unaware of whether any particular patient had received vitamin C.

Table I presents details concerning the 108 cases of influenza.

Table I.

Group (A) (400 mg. of vitamin C)					Group (B) (Controls)				
Case No.	Age in yrs	Initial Temp. (°F)	Duration of Illness in days	Sequelae, or Relapses	Case No.	Age in yrs	Initial Temp. (°F)	Duration of Illness in days	Sequelae, or Relapses.
1	21	98.6	8	-	2	20	99.	8	-
3	25	98.8	8	-	4	20	100.	8	-
5	42	97.6	8	-	6	32	98.8	8	-
7	52	98.8	8	-	8	19	99.	8	-
9	43	98.7	8	-	10	19	99.2	8	-
11	23	102.	8	-	12	25	98.8	10	-
13	39	99.	10	-	14	46	98.8	10	-
15	24	100.3	10	-	16	20	99.	10	-
17	24	99.1	10	-	18	31	97.6	10	-
19	42	97.6	10	-	20	20	99.2	12	-
21	21	99.1	10	-	22	50	99.2	12	-
23	21	99.2	10	-	24	46	99.1	12	-
25	43	100.	10	-	26	22	98.	11	Relapse
27	39	98.2	10	-	28	30	97.6	13	Relapse
29	19	98.6	12	Relapse	30	18	98.	11	Relapse
31	22	100.8	13	-	32	31	97.6	12	-
33	32	101.2	15	Relapse	34	27	99.	15	Relapse
35	24	98.2	9	-	36	39	98.	15	Relapse
37	50	98.	9	-	38	19	101.8	15	Gastritis
39	30	100.4	9	-	40	26	100.	9	-
41	37	98.4	9	-	42	20	101.	9	-
43	20	100.4	14	Relapse	44	34	103.	17	Relapse
45	21	98.4	10	-	46	30	97.6	10	-
47	23	100.	16	Relapse	48	26	98.	10	-
49	24	99.6	9	-	50	39	98.2	10	-
51	30	98.	9	-	52	20	99.2	14	Relapse
53	49	99.2	9	-	54	30	98.6	15	Sinusitis
55	21	100.2	31	Pneumonia	56	19	100.8	20	Gastritis
57	34	99.6	31	Bronchitis	58	19	97.6	19	Pneumonia
59	30	100.8	10	-	60	23	100.2	13	-
61	25	98.6	7	-	62	35	98.	23	Otitis
63	18	102.	7	-	64	53	100.2	41	Pneumonia
65	42	97.6	8	-	66	20	99.8	8	-
67	21	97.8	8	-	68	17	98.	10	-
69	28	98.	8	-	70	29	99.	8	-
71	45	98.3	13	-	72	38	97.6	10	-

(continued on next page)

Table I (continued).

Group (A)					Group (B)				
No.	Age	Temp.	Duration	Relapses	No.	Age	Temp.	Duration	Relapses.
73	39	100.	8	-	74	34	99.	11	-
75	28	100.2	12	Bronchitis	76	29	97.6	9	-
77	27	98.	8	-	78	35	97.9	9	-
79	31	100.	10	-	80	24	103.	12	Bronchitis
81	30	100.4	8	-	82	19	99.4	7	-
83	26	99.	11	-	84	19	98.8	7	-
85	23	99.4	6	-	86	44	98.	15	Relapse
87	21	101.2	8	-	88	26	101.	16	Relapse
89	23	99.4	10	-	90	26	97.6	8	-
91	29	98.2	7	-	92	19	98.	7	-
93	37	98.6	7	-	94	38	97.8	12	-
95	23	98.	7	-	96	32	97.6	10	-
97	39	99.	9	-	98	27	100.	8	-
99	40	100.8	9	-	100	32	104.8	13	-
101	30	97.6	8	-	102	24	98.	10	-
103	41	98.2	9	-	104	29	98.2	8	-
105	39	97.6	8	-	106	34	98.	12	-
107	20	98.5	12	-	108	42	98.4	8	-

Comparability of the Groups.

The two series, vitamin C cases and controls, are similar in respect of occupation, previous diet, etc., but there are two points in which they are not necessarily identical: a larger number of initially severe cases might have chanced to fall into one series; and the proportion of elderly men - who might be expected to recover more slowly - might not be equal in the two series.

Perhaps the best single criterion of initial severity is the degree of pyrexia at the time of admission to hospital. Judged by this standard the groups are similar, with a very slight bias in favour of the controls:

Initial Temperature	Number of patients of Group (A)	Group (B)
98.5° and under	19	24
98.6 - 99.5°	16	17
99.6 -100.5°	12	6
100.6° and over	7	7
	54	54.

An attempt to divide the patients, when first examined, into Mild, Moderate and Severe cases shows even less disparity between the groups:

	Group (A)	Group (B)
Number of mild cases	11	14
" " moderate "	32	31
" " severe "	11	9

As for age-distribution, there is a slight preponderance of/

of older men in the vitamin C group and of young lads in the control group:

Age.	Number of patients from Group (A)	Group (B).
17 - 21 years.	11	17
22 - 39 "	32	31
40 years and over	11	6

However, study of the individual cases shows that there is no justification for the view that older men would recover more slowly: of the 11 older men in Group (A), not a single one had a relapse or developed a complic--ation, and the average duration of their illness was actually a little less than the average duration in the whole group. Equally, there is no evidence to suggest that young lads were more severely affected: in the 17 youths of Group (B) the incidence of complications or relapses (29.4%) is almost identical with the incidence in the entire group (29.6%); and the average duration of illness among the youths was a shade less than the average for the whole group. Moreover, the average duration of illness for each series of 54 patients (10.2 days for the vitamin C cases and 11.78 days for the controls) corresponds very closely with the average for the 22 - 39 years age-group (10.0 days for the vitamin C cases and 11.58 days for the controls).

The two groups, then, may legitimately be compared.

Comparison of the Groups in respect of Complications and Relapses.

In the 54 cases treated with vitamin C there were 4 relapses and 3 complications (one case of pneumonia and two of bronchitis); of the 54 controls 9 had relapses and 7 developed complications (two cases of pneumonia, two of gastritis, and one each of otitis media, sinusitis and bronchitis).

The probability of an uncomplicated convalescence was therefore 87.04 % in the vitamin C cases and 70.37 % in the controls. The standard error of the probabilities

$$\text{is } \sqrt{\frac{87.04 \times 12.96}{54} + \frac{70.37 \times 29.63}{54}} \quad \text{or} \quad \pm 7.71 .$$

Hence the actual difference between the probabilities, 16.67 %, is 2.16 times the standard error.

Comparison /

Comparison of the Groups in respect of Duration of Illness.

The main points can be summarised thus:-

Duration of illness in days.	Number of patients from Group (A)	Group (B).
6	1	-
7	5	3
8	15	11
9	10	4
10	12	11
11	1	3
12	3	7
13	2	3
14	1	1
15	1	5
16	1	1
17	-	1
19	-	1
20	-	1
23	-	1
31	2	-
41	-	1
	54	54
Total days of illness	551	636
Average duration of illness	10.2 days	11.78 days.
Median " " "	9 "	10 "
Lower Quartile	10 "	13 "
σ	± 4.56	± 5.27

Standard error of difference = ±0.95
Actual difference of means = 1.58, or
1.66 times standard error.

Discussion.

In the 54 cases of influenza treated with ascorbic acid the incidence of complications and relapses was lower, and the average duration of illness was less, than in the 54 controls; but the differences were a little below the lower limit acceptable for statistical signif--icance. The experiment, then, does not permit of the drawing of definite conclusions. The probability is that oral administration of 400 mg. of vitamin C to a case of a fairly mild type of influenza does shorten the illness and reduce the risk of complications, but there is a possibility that the apparent beneficial results of vitamin C therapy are really due to chance.

The dosage of vitamin C employed was based on Falke's finding that an additional 100 mg. were required daily during pyrexia. As patients were admitted to hospital within 48 hours of the onset of their illness, and as observation of earlier cases (not included in the series) had shown that they remained febrile for - on the average - about 2 days after admission, a dosage of 400 mg. in two days/

days was selected. It was felt, however, that an attempt should be made to ascertain whether the dosage given was really adequate. For this purpose plasma and blood estim--ations seemed unsatisfactory, since - as has been pointed out earlier - ingestion of large amounts of the vitamin may cause a temporary rise in the blood concentration at a time when the tissues are still partially depleted; the use of a saturation test was ruled out by the nature of the experiment; and Meiklejohn and Stewart had not yet devised their enzyme test . In default, therefore, of a better method, the dichlorophenol-indophenol test of urinary excretion was employed.

Excretion of Vitamin C in Influenza.

(a) Controls. The urinary concentration and total excretion of 32 of the control cases was estimated during their first three days in hospital. In spite of the fact that these patients received a little aspirin, a drug that increases the urinary output of ascorbic acid, the output was uniformly low, as the following summary will reveal:

Concentration in mg./cc.	Number of patients. 1st. day.	2nd. day.	3rd. day.
Under .005	6	7	7
.005 -.006	12	11	10
.006 -.009	10	11	10
Over .009	4	3	3
	32	32	30 *
Total excretion in mg.			
Under 5	10	11	12
5. - 6	6	6	6
6.1 - 10	8	7	7
Over 10	8	8	5
	32	32	30 *

* Two specimens lost.

The highest concentration recorded for any patient on any of the three days was .0099 mg./cc., a dangerously low concentration. The lowest concentration was .0037 mg./cc., and the average was .0064, with σ of 0.20. The highest total excretion on any day was 14.4 mg., the smallest was 3.6 mg., and the mean was 7.24 mg.

(b) Cases given 400 mg. of vitamin C. The urinary concentration of 44 of the treated cases was estimated after one complete day in hospital, - i.e. after they had received 200 mg. of ascorbic acid. The concentration was found to range from .003 to .0125 mg./cc., with a mean of .0083 mg/cc. The total excretion during the first complete day in hosp--ital varied between 4.6 and 19.2 mg, with a mean of 8.82 mg.

The estimations were repeated on 24 of the patients after/

after they had received the full 400 mg. The concentration now ranged from .0071 to .015 mg./cc., with a mean of .0111 mg./cc.; and the total excretion ranged from 9.9 to 22.4 mg., with an average of 16.6 mg.

It appears, then, that the vitamin C excretion of the controls was very low indeed; that the first 200 mg. of the vitamin given to the treated cases was practically all retained by the tissues, and caused very little increase in the excretion; and that the second 200 mg. resulted in an appreciable rise in the excretion, but that even this rise did not bring the excretion to the level of normality.

These findings made it clear that 400 mg. of ascorbic acid were insufficient to remedy the state of hypovitaminosis present in influenza. Since influenza is not often treated in a hospital for infectious diseases, no opportunity occurred for repeating the experiment with larger dosage of the vitamin. For subsequent experiments on other infections, however, a larger dosage was employed.

(2) CEREBROSPINAL MENINGITIS.

The 1940 epidemic of cerebrospinal fever afforded opportunity for the investigation on a considerable scale of the possible effects of ascorbic acid therapy. In view both of the rapidity which often characterises a toxic case of " spotted fever " and of the prevalence of symptoms of gastro-intestinal upset (one of the undesirable effects of sulphapyridine), it was deemed desirable to investigate separately the effects of oral and parenteral administration of the vitamin.

Experiment I - Oral administration of vitamin C.

For this investigation 120 consecutively admitted cases of cerebrospinal meningitis were divided - on the alternate case principle - into two groups of 60. Members of Group (A) - i.e. every alternate case admitted during the period - were offered vitamin C in tablet form for four days, the dosage for adults being 50 mg. thrice daily (i.e. 600 mg. in all), while children between the ages of two and twelve years were given 50 mg. twice daily (i.e. 400 mg. in all), and infants under two years received 25 mg. twice daily (or 200 mg. in all). Members of Group (B) were given no supplement of vitamin C, but, in all other respects, received the same treatment (standard dosage of sulphapyridine, etc.) and the same diet.

In all cases the criterion of diagnosis was the presence of Gram negative diplococci in the cerebrospinal fluid of a patient who showed clinical signs of meningitis.

As many of the patients in Group (A) were too sick to tolerate vitamin C given by mouth, and as exclusion of these patients without any corresponding exclusions from Group (B) would obviously invalidate any conclusions, it is necessary to select a line of demarcation equally applicable to both groups. Since ability to tolerate sulphapyridine by the oral route obviously implies ability to retain vitamin C given by mouth (although the converse is not true), it seems to be the obvious choice for the line of demarcation.

The patients excluded on the ground of inability to tolerate sulphapyridine given by the oral route include 18 from Group (A) and 14 from Group (B); we are therefore left with 42 cases treated with ascorbic acid, and 46 controls.

Details of individual cases are given in tables II and III.

Table II.

Cases of meningitis treated with ascorbic acid.

No.	Sex.	Age	Complications.	Duration of illness.
1	F	17 yrs.	-	27 days
2	M	8	Arthritis, transient	30 "
3	M	18	-	29 "
4	M	8/12	Died 44 hrs. after admission.	
5	M	23	-	28 days
6	M	1 6/12	-	36 "
7	M	9/12	-	32 "
8	F	56	Died 74 hrs. after admission.	
9	M	3	Ataxia, transient	54 days
10	M	11	-	36 "
11	M	30	-	28 "
12	F	28	Arthritis, transient	38 "
13	M	1 7/12	-	44 " *
14	M	24	-	29 "
15	M	10	Leg pains, lasted	29 "
16	F	28	-	30 "
17	M	28	-	27 "
18	F	5	-	30 "
19	F	5/12	-	29 "
20	M	21	-	44 " °
21	F	37	Arthritis, transient	53 "
22	F	26	-	46 "
23	F	12	-	27 "
24	M	1 1/12	-	32 "
25	M	11/12	-	29 "
26	F	30	-	29 "
27	M	7	Ataxia, lasted	27 "
28	M	6	-	27 "
29	M	21	-	29 "

(Continued on next page)

* Discharge delayed through intercurrent varicella.
° " " " " tonsillitis.

Table II (continued).

No.	Sex.	Age in years.	Complications.	Duration of illness in days
30	M	50	-	28
31	M	1 9/12	-	27
32	M	20	-	30
33	F	22	-	31
34	M	24	Leg pains, lasted	29
35	F	7/12	-	27
36	M	3	-	36
37	F	18	-	28
38	M	27	-	29
39	M	17	-	28
40	M	33	Ataxia, lasted	27
41	M	38	-	26
42	F	6	Arthritis, transient	28 .

Table III.
Cases of meningitis used as controls.

No.	Sex.	Age in years.	Complications.	Duration of illness in days.
1	F	16	-	24
2	M	54	Leg pains, lasted	33
3	M	4/12	Nuchal rigidity, lasted*	27
4	F	4	-	28
5	F	9/12	-	41
6	M	20	Ataxia, lasted	28
7	M	6	-	28
8	M	20	Ataxia, lasted	28
9	M	1 7/12	-	28
10	F	18	-	26
11	M	3	-	27
12	M	3	Ataxia, transient	29
13	M	17	-	27
14	M	5	Otitis, transient	43
15	F	9/12	-	42 °
16	M	30	-	26
17	F	1 4/12	Relapse, transient	37
18	F	2	Died on 64th. day (hydrocephalus)	
19	F	12	-	30
20	M	18	Tachycardia, lasted	47
21	F	16	Ataxia, transient	29
22	F	66	-	28
23	M	16	-	27
24	M	1 8/12	-	35
25	M	27	Arthritis, lasted	38
26	M	20	-	32
27	F	19	-	29
28	M	1	-	28
29	M	1 7/12	-	29

(Continued on next page)

* Died later of another disease; post mortem showed old meningeal adhesions

° Discharge postponed because of intercurrent varicella.

Table III (continued).
Cases of meningitis used as controls.

No.	Sex.	Age in years.	Complications.	Duration of illness, in days.
30	F	3/12	Relapse, transient	44
31	M	2 9/12	-	29
32	M	65	Died 59 hrs. after admission.	
33	M	3/12	-	31
34	F	3/12	Died on 5th. day in hospital.	
35	M	5	-	29
36	M	2 2/12	-	28
37	M	31	-	26
38	F	9/365	-	34
39	M	25	-	25
40	M	9	-	28
41	F	13	-	27
42	M	52	Died on 6th. day in hospital.	
43	F	4	Died 16 hrs. after admission.	
44	F	3	Died 23 hrs. after admission.	
45	M	44	Arthritis, lasted	44
46	M	2	Died on 10th. day in hospital.	

(N.B. The word "lasting", as used in tables II and III, means that the particular complication or sequel so described was still present when the patient left hospital; it does not necessarily imply permanence.)

Comparison of the Groups.

Some of the main points regarding the two groups may be summarised thus:-

	Group (A) (Vitamin C)	Group (B) (Controls)
1. Total patients.		
No. of cases	42	46
Deaths	2 (4.76%)	7 (15.22%)
Lasting sequelae	4 (9.52%)	7 (15.22%)
Transient complications	5 (11.9 %)	5 (10.87%)
Recoveries without "	31 (73.81%)	27 (58.7 %)
Mean duration in survivers	31.2 days	30.8 days.
2. Patients aged less than three years.		
No. of cases	9	16
Deaths	1 (11.11%)	3 (18.75%)
Lasting sequelae	0	1 (6.25%)
Transient complications	0	2 (12.5 %)
Recoveries without "	8 (88.89%)	10 (62.5 %)
3. Patients aged more than 45 years.		
No. of cases	2	4
Deaths	1 (50 %)	2 (50 %)
Lasting sequelae	0	1 (25 %)
Recoveries without complic'n.	1 (50%)	1 (25 %).

4. Patients aged 3 - 45 years (inclusive).	Group (A).	Group (B).
Total cases	31	26
Deaths	0	2 (7.69%)
Lasting sequelae	4 (12.9 %)	5 (19.23%)
Transient complications	5 (16.13%)	3 (11.54%)
Recoveries without "	22 (70.97%)	16 (61.54%).

A direct comparison of the 42 patients of Group (A) with the 46 controls of Group (B) would give the erroneous impression that, while the average duration of illness in survivers is almost identical in the two groups and while the difference in incidence of complications is slight, there is a difference in the probability of death which, although not actually statistically significant (it is 1.68 times the standard error of the difference), is yet highly suggestive. Such an impression would be quite unjustified, because the control group is handicapped by its unduly large proportion of infants and elderly persons, - the very sections of the community in which the vast majority of deaths from meningococcal meningitis occur.

Since the disparity of the age-distributions makes direct comparison of the two groups utterly valueless, we are compelled either to compare the two series age-group by age-group (in which case the differences in incidence of sequelae and in percentage of deaths will be found to be constantly in favour of the patients who received vitamin C. but will also be found to be invariably very far below the level of significance), or to try to obtain a picture of the real differences between the groups by standardising the death rates and complication rates.

Since it is immaterial what population we take as our standard, it is convenient to take the age-distribution of Group (B) as the standard - so that the rates for that Group need no alteration - and to calculate the rates that would obtain in Group (A) if the number of patients in each age-group were raised or lowered to that of the corresponding age-group of Group (B). Standardisation reveals that, although the differences are certainly in favour of Group (A), these differences are very far from being significant:-

Standardised death rate of Group (A) = 8.21%.
" " " " " (B) = 15.22%.
Difference = 7.01%, or 1.05 times the standard error of the difference (±6.67)

Standardised complicatn rate of Group (A)= 16.43%.
" " " " " (B)= 26.09%.
Difference = 10.34%, or 1.22 times the standard error of the difference (±8.47).

Experiment /

Experiment II (a) - Parenteral administration of vitamin C to toxic cases of cerebrospinal meningitis.

While Experiment I (on oral therapy with ascorbic acid) was in progress it was felt that some of the patients who were unable to tolerate drugs by mouth might be utilised for investigation of the effects of parenteral administration of the vitamin. Accordingly, all those patients of the original Group (A) who - after 24 hours in hospital - were still unable to tolerate sulphapyridine orally were given an intramuscular injection of 500 mg. of ascorbic acid, approximately 24 hours after admission to hospital; and those of the Group (B) patients who, after a similar lapse of time, still required parenteral administration of sulphapyridine were regarded as controls.

The 500 mg. of the vitamin given intramuscularly were, of course, additional to any portion of the first day's oral dosage that the patients managed to retain.

As was stated earlier, 32 out of 120 cases had to be excluded from Experiment I on the ground of inability to tolerate drugs by mouth. The first 7 of these 32 were either convalescent or dead before Experiment II (a) was commenced. The remaining 25 - twelve from Group (A) and thirteen from Group (B) - formed the material for the experiment.

Twenty-five cases are too few to permit of the drawing of conclusions, but - as these cases will be used later in conjunction with the cases of Experiment II (b) - details of the 25 are given in tables IV and V.

Table IV.

Toxic cases of meningitis given vitamin C parenterally.

No.	Sex.	Age in yrs.	Rash?	Grade*	Complications.	Duration of illness in days.
1	F	30	Yes	+++	Neuritis, lasted	30
2	M	23	No	+++	-	34
3	M	63	No	+++	Died 61 hrs. after admission.	
4	M	44	Yes	+++	-	30
5	M	30	No	++	-	30
6	M	14	Yes	+++	-	29
7	M	25	Yes	+++	-	27
8	F	4½	No	+	Died 55 hrs. after admission.	
9	M	15	No	++	-	27
10	F	6	Yes	++++	Died 32 hrs. after admission.	
11	F	35	Yes	+++	Neuritis, lasted	23°
12	M	6	No	+++	-	31

Table /

° Patient left on her own responsibility.

* Grades of initial severity: + = Mild; ++ = Average; +++ = Severe; ++++ = Moribund.

Table V.

Toxic cases of meningitis used as controls.

No.	Sex.	Age in years.	Rash?	Grade.	Complications.	Duration of illness in days.
1	M	5	Yes	+++	Died 65 hrs. after admission.	
2	F	72	No	++	Died 61 hrs. after admission.	
3	F	30	No	+	Neuritis, transient	28
4	M	2½	No	+++	Died 53 hrs. after admission.	
5	M	34	No	++	Arthritis, transient	28
6	M	30	No	++	-	26
7	M	32	No	+++	-	27
8	F	31	No	+++	-	30
9	F	37	No	++	-	28
10	M	4	Yes	+++	Died 64 hrs. after admission.	
11	M	4/12	No	++++	Died 45 hrs. after admission.	
12	M	18	Yes	+++	Deafness, lasted	44
13	M	28	Yes	+++	Died 40 hrs. after admission.	

Comparison of Groups.

Some of the main points of difference between the two groups may be expressed thus:-

	Toxic cases treated with vitamin C.	Toxic Controls.
Total cases	12	13
Deaths	3 (25. %)	6 (46.15%)
Complications	2 (16.67%)	3 (23.08%)
Recoveries without any complication	7 (58.33%)	4 (30.77%)
Average duration of illness in survivers	29.75 days.	30.14 days.

To dispose in advance of any suggestion that the mortality of the control group was so unusually high as to render that group unsuitable for use in a comparison, it must be emphasised that these twenty-five patients were not ordinary cases of cerebrospinal meningitis, but cases selected on the ground of being too ill to tolerate sulphapyridine orally, - i.e. selected from the group in which the majority of deaths occur.

No patient in these two groups was able to take sulphapyridine by mouth; a petechial rash (which is a rough indication of extent of septicaemia) was present in six of the vitamin C cases as compared with four of the controls; clinical division of the patients (by the writer) into various grades of initial severity suggested that the vitamin C cases were, on the average, a shade more toxic on admission; and there was little difference in the age-distributions of the two groups. In these circumstances it was felt that the reduction in mortality rate in the vitamin C cases was - by comparison with the controls - sufficiently marked to make a further investig-ation of the effects of parenterally administered ascorbic acid/

acid eminently desirable.

Experiment II (b) - Parenteral administration of vitamin C.

The experiments previously described were performed on one hundred and twenty consecutive cases of cerebrospinal meningitis. For the next thirty cases admitted to hospital it was decided to give each alternate patient ascorbic acid in large amounts, intramuscularly, the dosage chosen being 500 mg. daily for three days for an adult or a child over the age of five years, and half that daily quantity for younger children. Unfortunately, two cases of pneumococcal mening-itis were included in error, - a result of treatment being started as soon as lumbar puncture had revealed a turbid cerebrospinal fluid.

Details of individual cases are given in tables VI and VII.

Table VI.

Cases of meningitis given vitamin C parenterally.

No.	Sex.	Age in years.	Rash?	Grade.	Complications.	Duration of illness in days.
1	M	5	Yes	+++	-	28
2	F	8	Yes	+++	-	28
3	M	18	Yes	++++	-	31
4	(Pneumococcal, - excluded.)					
5	M	5/12	Yes	++	-	28
6	F	25	No	+++	-	26
7	F	9/12	No	+++	-	26
8	M	4	No	++	-	27
9	M	20	Yes	++	-	27
10	M	14	Yes	+++	-	28
11	F	14	No	+++	-	32
12	(Pneumococcal, - excluded.)					
13	M	21	No	++	-	27
14	F	1	No	++	-	40
15	M	16	No	+++	-	30

Table VII.

Cases of meningitis used as controls.

No.	Sex.	Age in years.	Rash?	Grade.	Complications.	Duration of illness in days.
1.	M	27	No	+++	Neuritis, lasting; & Haematuria, transient	44
2.	F	6/12	No	++	-	41
3.	M	2	Yes	+++	-	30
4.	F.	42	No	+++	Died 70 hrs. after admission.	
5.	F.	17	No	++	-	28
6.	M.	14	Yes	++	-	28
7.	M.	3	Yes	+++	Arthritis, lasting	25*
8.	F.	55	Yes	+++	Neuritis, transient	26
9.	F.	14	No	++	-	25

(Continued on next page.)

* Transferred to another hospital for treatment of arthritis.

Table VII (continued).

No.	Sex.	Age in years.	Rash?	Grade.	Complications.	Duration of illness in days.
10	M	2	Yes	+++	-	36
11	M	5/12	No	++	-	31
12	F	20	No	++	-	25
13	M	52	Yes	+++	Died on 6th. day in hospital.	
14	M	5	No	++	-	30
15	M	6/12	No	++++	Died 10 hrs. after admission.	

The outstanding feature of these cases is, of course, the absence of any deaths, complications, or sequelae in the group treated with intramuscular injections of ascorbic acid.

Comparability of the Groups.

An attempt by the writer to classify the patients immediately after admission into various grades of initial severity showed little disparity between the two groups: one member of each group was regarded as " moribund", seven of each group were classed as " severe", while five vitamin C cases and seven controls were judged to be " moderate". Rashes were present in six members of each group. The sex distribution was similar, - 8 males and 5 females in the vitamin C group, and 9 males and 6 females in the controls. Apart from the fact that the control group contained two patients in late middle-age, the age-distributions were much the same in the two series:

Age in years.	Vitamin C cases.	Controls.
Under 2	3	4
2 - 10	3	3
11 - 20	5	4
21 - 39	2	2
40 - 55	0	2
	13.	15.

Comparison of the Groups.

The two series of cases are here compared in respect of deaths, complications, and duration of illness. With regard to the last of these points, it should be mentioned that the discharging of the patients was in the hands not of the writer but of a colleague who did not know to which group any particular patient belonged.

The main features may be summarised thus:-

	Vitamin C cases.	Controls.
Total cases	13	15
Deaths	0	3 (20%)
Complications	0	3 (20%)
Recoveries without complications	13	9 (60%)
Average duration of illness in survivers	29.08 days	31.27 days.

(In calculating the average duration of illness, the patient -

Control /

Control Group, No. 7 - who was transferred to another hosp--ital for treatment of a complication, has been excluded.)

It was hardly to be expected that such small numbers would provide differences large enough to be statistically significant. Yet the standard error of the probabilities of recovery without complications is 12.65, so that the actual difference - 40 % - is more than three times the standard error.

If we add together the figures for Experiments II (a) and II (b), we have:

	Parenteral vitamin C cases.	Controls.
Total cases	25	28
Deaths	3 (12. %)	9 (32.14%)
Complications	2 (8. %)	6 (21.43%)
Recoveries without complications	20 (80. %)	13 (46.43%).

The difference between the rates for recoveries without any complication is 33.57 %, or 2.714 times the standard error of ± 12.37.

As for the death rates, the actual difference is a fraction under twice the standard error, but it is obvious that, in Experiment II (a), where the ascorbic acid was not given until the patient had been in hospital for 24 hours, it could not reasonably be expected to prevent the death of one patient who expired early on her second day in hospital. Exclusion of all patients who died within 36 hours of their admission would give a death rate for the vitamin C cases of 2/24, or 8.33 %, as against 8/27, or 29.63 %, for the controls; and the difference - 21.3 % - is well over twice the standard error.

Equally informative is a division of these 53 cases into age-groups:

	Ratio of deaths to cases.		Ratio of complications to survivers.	
Age.	Vitamin C cases.	Controls.	Vit. C cases.	Controls.
Under 1 year	0/2	2/4	0/2	0/2
1 - 3 years	0/1	1/4	0/1	1/3
4 - 19 "	2/12	2/7	0/10	1/5
20 - 39 "	0/8	1/9	2/8	3/8
40 years and over	1/2	3/4	0/1	1/1
Total	3/25	9/28	2/22	6/19.

In not a single age-group does the advantage rest with the controls.

Comparison of the entire 148 cases of meningitis.

The entire 148 cases can be classified into five groups as follows/

follows:-
(a) Early controls, - i.e. the 60 patients who were originally taken as controls for Experiment I. (46 of these were actually used as controls for Experiment I, the remaining 14 being too sick to tolerate drugs orally; and the last 13 of these sick patients were used as controls for Experiment II (a).)
(b) Late controls, - i.e. the 15 patients used as controls for Experiment II (b). (The 75 controls represent every alternate case admitted during the period of the experiments).
(c) Early " oral " cases, - i.e. patients who were offered ascorbic acid by mouth and were not given any ascorbic acid parenterally. (This group includes all of the 60 cases originally taken for Experiment I, except the 12 who subsequently received vitamin C intramuscularly. For comparative purposes this group is therefore in a position of favourable bias, in that 12 very sick patients have been excluded from it and placed in a separate category.)
(d) 500 mg. group, - i.e. the 12 toxic cases, too sick to tolerate sulphapyridine orally, that received single injections of vitamin C. (For comparative purposes this group is under a handicap: it consisted entirely of selected toxic cases.)
(e) 1500 mg. group, - i.e. the 13 cases used in Experiment II (b).

The five groups can be summarised thus:-

Group.	No. in group.	Died within 36 hrs.	Died later.	Developed complications.	Recovered without complications developing.
(a) Early controls.	60	2	11	15	32 (53.33%)
(b) Late controls.	15	1	2	3	9 (60. %)
(c) Early " oral " cases.	48	3	5	9	31 (64.58%)
(d) 500 mg. group.	12	1	2	2	7 (58.33%)
(e)1500 mg. group.	13	0	0	0	13 (100. %)
Total.	148	7	20	29	92 (62.16%).

In the early controls and the late controls the death rates are almost identical (21.67 % and 20. % respectively), and the incidence of complications does not differ greatly. It appears, then, that during the portion of the epidemic under consideration the disease did not undergo any marked change in its severity. Hence there is no valid reason why the Group (e) cases should not be compared with the whole 75 controls. The comparison will /

will be made in respect of mortality rates, incidence of complications, and percentage of recoveries without any complication.

(1) Death rates. Among the seventy-five controls the probability of dying is 21.3 %, with a standard error of ± 4.56 . In the patients who received 1500 mg. of vit--amin C the probability of dying is 0 %. The difference is, therefore, definitely significant. Even if we exclude the three controls who died early (on the ground that no drug could benefit such cases), the difference between the mortality rates of the 1500 mg. patients and of the controls remains statistically significant.

(2) Incidence of complications. Among the controls the incidence of complications is 24 %, with a standard error of ± 4.93 . Again, the difference in the 1500 group - where the incidence is 0 % - is significant.

(3) Probability of recovery without any complications. In the controls the probability of an uncomplicated conval--escence is 54.67 %, with a standard error of ± 5.75. The probability in the 1500 mg. group is 100 %, and the differ--ence is definitely significant.

Finally, let us compare the incidence of uncomplicated convalescences in the 25 patients who received vitamin C parenterally and in the other 123 patients. The dice, in this comparison, are weighted against the 25 parenteral cases, because twelve of them (the Group (d) cases) were not unselected cases, but toxic cases unable to take drugs orally. The incidence of uncomplicated recoveries in the 123 patients who received no ascorbic acid parenterally is 58.54 %, and the corresponding rate in the 25 patients who received one or more injections of the vitamin is 80 %. The standard error of the difference is

$$\sqrt{\frac{80 \times 20}{25} + \frac{58.54 \times 41.46}{123}}$$ or ± 9.15 . So the actual difference, 21.46 %, is - even in this weighted comparison - more than twice the standard error.

Every single comparison, then, emphasises the marked differences between cases treated with intramuscular injections of vitamin C and controls who received no such injections.

Experiment III - Confirmatory.

A brief confirmatory experiment was conducted at Kendray Isolation Hospital, Barnsley, every alternate case admitted/

admitted with cerebrospinal meningitis receiving daily injections of 500 mg. of ascorbic acid during the first three days in hospital. Except in case No. 8, the first injection was always given intravenously; the second and third injections in all cases, and the first injection in case 8, were given intramuscularly. The criterion of diagnosis and the general treatment were exactly the same as in the Edinburgh cases.

Details of individual cases are given in table VIII, but " Duration of illness " is deliberately omitted, because the discharging of the patients was in the hands of the writer who might be guilty of unconscious bias.

Table VIII.

Cases of meningitis used in confirmatory experiment.

No.	Dosage of vit. C.	Sex.	Age in years.	Rash?	Grade.	Complications.
A1	1500 mg.	F	11	No	++	-
A2	1500 mg.	M	13	No	+++	-
A3	1000 mg.	M	2	No	+	-
A4	1500 mg.	F	39	Yes	+++	-
A5	1500 mg.	F	7	No	+++	Strabismus, transient.
A6	1500 mg.	F	21	No	+++	-
A7	1500 mg.	F	36	Yes	+++	Haematuria, transient.
A8	1000 mg.	M	1 1/12	Yes	++	-
B1	Nil.	M	29	No	++	Otitis, transient.
B2	"	M	5	Yes	++	Neuritis, lasted.
B3	"	M	5	Yes	+++	-
B4	"	M	1 1/12	Yes	++	-
B5	"	M	4/12	No	+++	Died on 7th. day in hospital.
B6	"	M	20	No	+++	Deafness, lasted.
B7	"	F	28	Yes	++	Deafness, lasted.
B8	"	M	1 9/12	No	+	-

Comparison of the groups.

These two short series, comparable enough in respect of initial severity, are - although Group B contains one infant appreciably younger than the youngest child in Group A - not dissimilar in age-distribution. In the absence of any evidence that the meningococcus is more toxic to males (who predominate in Group B) than to females, the groups may legitimately be compared.

The main points of difference are:-

	Vitamin C cases.	Controls.
Total cases	8	8
Deaths	0	1
Transient complications	2	1
Lasting sequelae	0	3
Recoveries without any complications	6	3 .

These/

These cases are not sufficiently numerous to be significant by themselves: for example, the difference in the probabilities of uncomplicated convalescence is only 1.63 times the standard error of the difference. Since, however, the observed results in these cases accord with the findings obtained in the Edinburgh cases, they afford very valuable corroborative evidence.

Their value does not lie in the fact that the ascorbic acid cases and the controls can be grouped with their respective Edinburgh counterparts to form two series which differ significantly. It lies, rather, in the fact that the presence of similar findings in a second series of cases virtually eliminates the possibility of chance playing a part. So long as we have only one series of cases and controls, the differences - even though they satisfy all the criteria of statistical significance - might conceivably be due to some freak of chance: after all, the generally accepted criterion of a " significant " difference - a magnitude greater than two and a half times σ - simply means that there is less than one chance in a hundred that the difference is coincidental. But it would be in the nature of a miracle if the very freak of chance that occurred in one series were to be repeated in the next series. - To adopt the parlance of the race-course, if the odds against the Edinburgh results (considered by themselves) being due to chance are estimated at about 150 to 1, and if the odds against the Barnsley results (considered alone) being due to chance are estimated at about 75 to 1, then the odds against the results of the two series (considered together) being due to chance will be in the neighbourhood of 11,000 to 1.

Conclusions.

The value of parenteral administration of vitamin C as an adjuvant to the sulphapyridine treatment of cerebrospinal meningitis is proved beyond shadow of doubt. Cases so treated showed a very marked decline, as compared with appropriate controls, both in the percentage of deaths and in the incidence of complications and sequelae.

On the other hand, oral administration of the vitamin is often impracticable owing to the alimentary upset induced by sulphapyridine; and even in those cases of meningitis that can tolerate oral medication the evidence in favour of administration of ascorbic acid by mouth is very indefinite: cases so treated appeared to show a slight reduction in the frequency of complications and in the average duration of illness, but these apparent reductions were so slight that a very extensive experiment would be necessary before it could be ascertained whether they were significant or due to chance.

(3)/

(3) STREPTOCOCCAL TONSILLITIS AND SCARLET FEVER.

(a) Tonsillitis.

During the investigation of ascorbic acid therapy in diphtheria (*vide infra*), a number of patients, who were at first placed either in the group of " Mild cases of diphtheria treated with vitamin C " or in the group of " Mild cases of diphtheria used as controls ", had to be excluded after laboratory examination had revealed that the only pathogenic organism present in throat swabs was Str. Pyogenes. As these cases accumulated, it was felt that they might be used for an investigation of the effects of vitamin C in tonsillitis.

Accordingly, 32 cases of streptococcal tonsillitis were studied, the criterion of diagnosis accepted being an inflamed throat from which haemolytic streptococci were cultured. Some of these 32 cases were, as mentioned above, originally classed as " diphtheria ", while others were recognised from the beginning as tonsillitis. Such of the patients as originally fell into the category of "Mild cases of diphtheria treated with vitamin C " had invariably begun to receive ascorbic acid before they were recognised as cases of tonsillitis. Hence the use of the alternate case method was impracticable.

In all, 16 unselected cases were studied as controls, and 16 unselected cases were treated with vitamin C, the dosage employed being 100 mg. daily for eight days for adults and children over the age of eight years, and 50 mg. daily for younger children. Since none of the cases developed complications, comparison was limited to two points, - the duration of tonsillar inflammation, and the duration of stay in hospital. (" Duration of stay in hospital " was chosen, rather than " duration of illness ", because with young children the history of onset tends to be unreliable.)

Details of the individual patients are given in tables IX and X.

Table IX.

Cases of streptococcal tonsillitis treated with vitamin C.

No.	Sex.	Age in years.	Duration of inflammation of tonsils, in days.	Length of stay in hospital, in days.
1	F	8	7	14
2	M	6	16	19
3	M	3	7	10
4	M	30	11	13
5	M	18	6	12
6	M	1	6	8
7	M	19	9	10
8	F	15	6	18

(Continued on next page.)

Table IX (continued).

No.	Sex.	Age in years.	Duration of inflammation of tonsils, in days.	Length of stay in hospital, in days.
9	F	2	10	21
10	M	4	7	16
11	M	18	6	12
12	F	15	17	21
13	M	9	9	14
14	F	14	7	15
15	F	2	10	17
16	M	23	10	16 .

Table X.

Cases of streptococcal tonsillitis used as controls.

No.	Sex.	Age in years.	Duration of inflammation of tonsils, in days.	Length of stay in hospital, in days.
1	M	22	12	23
2	F	27	7	12
3	M	2	17	20
4	F	44	7	10
5	F	20	7	16
6	M	9	7	10
7	M	4	12	15
8	F	30	8	17
9	F	6	3	8
10	F	18	10	13
11	M	25	10	15
12	M	6	21	26
13	F	3	14	23
14	M	9	10	15
15	M	17	10	17
16	M	6	12	16 .

The main points regarding the two groups may be summarised as follows:

	Vitamin C group.	Controls.
Median duration of inflammation	8 days.	10 days.
Average " " "	9.0 "	10.44 "
σ of " " "	$\pm$3.29	$\pm$ 4.23
Median " " hospitalisation	14.5 days.	15.5 days.
Average " " "	14.75 "	16.0 "
σ of " " "	$\pm$ 3.76	$\pm$ 4.86

Actual difference in average duration of inflammation = 1.44 days; and standard error of difference = $\pm$1.34

Actual difference in average duration of hospitalisation = 1.25 days; and S. E. Difference = $\pm$1.54

Hence, although the average duration was less in cases of tonsillitis treated with vitamin C than in comparable controls, the application of statistical methods shows that the /

the differences were far below the level of significance.

(b) Scarlet Fever.

Forty cases of scarlet fever were also investigated, every alternate case receiving 100 mg. of vitamin C daily for eight days. The experiment was discontinued after the fortieth case for the following reasons:-

1. Since the mild type of scarlatina that has been prevalent in recent years gives rise to remarkably few complications, a very large-scale (and expensive) exper--iment would be necessary to determine whether treatment with ascorbic acid does anything to reduce the incidence of complications; moreover, in any such experiment, it would be essential to eliminate such other variables as dosage of serum (if any) and of sulphonamides (if any).

2. Apart from the incidence of complications and the duration of the incidental tonsillitis, it is difficult to find criteria on which to judge the efficacy of any treatment for a self-limiting disease like mild scarlatina.

3. The findings in respect of the duration of the tonsillar inflammation were very similar to the results obtained in the experiment on simple tonsillitis.

No useful purpose would be served by recording details regarding the individual cases. The only point that seems worth stating is that, in two fairly comparable groups each containing 20 patients, the average duration of tonsillar inflammation was 7.12 days in the cases treated with vitamin C, and 8.21 days in the controls, and that (since the standard error of the difference is ± 1.22) the difference is not significant.

Conclusions from experiments on tonsillitis and scarlet fever.

In cases of streptococcal tonsillitis, with or without a rash, the duration of tonsillar inflammation was rather less in cases treated with ascorbic acid than in controls, but the difference was well below the level of significance. Since the vitamin C cases appeared to advantage in two separate series (tonsillitis cases and scarlet fever cases) it seems quite likely that the vitamin is of some slight benefit, but only by a repetition of the experiment on a very large scale would it be possible to determine whether the observed differences were significant. *

(4) /

* As already mentioned on page 15, Glazebrook and Thomson in 1942 carried out a large-scale experiment and concluded that administration of vitamin C reduced the duration of tonsillitis.

(4) POST-INFLUENZAL PNEUMONIA.

Every alternate case admitted with bronchopneumonia following influenza was given vitamin C orally, the dosage employed being 200 mg. daily for three days. Apart from ascorbic acid, the patients received the same treatment.

Details of individual cases are given in table XI.

Table XI.

Cases of post-influenzal pneumonia.

No.	Age in yrs.	Sex.	Total vit.C in mg.	Day of disease on which patient entered hospital.	Day of disease on which temp. fell to normal.	Day of disease on which resp. rate fell to 24 or less.	Complic--ations.	Duration of illness in days.
A1	50	M	600	4	Died on 8th. day of disease.			
A2	27	F	600	2	3	4	-	26
A3	35	M	600	3	5	10	-	23
A4	30	M	600	4	7	7	-	22
A5	62	F	600	8	9	14	-	48 *
A6	30	M	600	5	7	7	-	22
A7	38	F	600	6	7	14	-	24
B1	51	M	-	2	3	6	-	18
B2	50	M	-	4	7	14	-	34
B3	45	F	-	7	8	15	-	36
B4	41	M	-	3	6	8	-	17 °
B5	26	M	-	5	Died on 9th. day of illness.			
B6	41	F	-	5	8	13	-	36
B7	23	M	-	1	3	5	-	53 +

* Discharge delayed because of kaolin burn.
° Left hospital on own responsibility before clinically well.
+ Discharge delayed because of of ulcer at site of injection of sulphapyridine.

Lack of patients brought the experiment to a close before a sufficient number had been investigated. However, the cases studied, small though their number is, at least suggest that any benefits conferred by vitamin C therapy are not great:

	Vitamin C cases.	Controls.
Average duration of pyrexia	6.33 days.	5.83 days.
Median " " "	7. "	6.5 "
Average " of rapid respiration	9.33 "	10.17 "
Median " " " "	8.5 "	10.5 "
Average duration of hospitalisatn.	27.5 "	32.33 "
Median " " "	23.5 "	35. ".
Percentage of deaths	14.3	14.3

(5) /

(5) DIPHTHERIA.

Experiment I - Oral administration of vitamin C.

In Edinburgh City Hospital cases of faucial diphtheria are habitually divided by the medical superintendent into three groups, - F 1, or mild cases, with scanty membrane and practically no toxaemia (Dosage of antitoxin less than 16,000 units); F 2, or moderate cases, with membrane covering both tonsils, some adenitis, and some toxaemia (Dosage of serum about 16,000 to 30,000 units); and F 3, or severe cases, with membrane spreading on to the pillars of the fauces, marked adenitis and toxaemia (Dosage of serum, 50,000 or 100,000 units). Laryngeal cases are similarly divided into L 1, L 2, and L 3, Nasopharyngeal cases into NP 1, NP 2 and NP 3, and the same procedure is adopted for the other forms of the disease.

It was felt that a fairer picture of the effects of ascorbic acid therapy would be obtained by applying the alternate case principle to each of these three classes - mild cases, moderate cases, and severe cases - than by simply giving vitamin C to every second case of diphtheria admitted to hospital. Accordingly, for a period of about five months, every alternate admission in each class was given ascorbic acid by mouth, the dosage selected being 100 mg. daily for eight days for patients of eight years or over, and 50 mg. daily for eight days for younger children. Out of 120 cases admitted to hospital, 116 were originally included in the experiment, the four exclusions being patients who died within a few hours of admission.

Subsequent exclusions on bacteriological grounds.

In 20 of the mild cases and 6 of the moderate cases the diagnosis of diphtheria was not confirmed bacteriolog--ically. These 26 cases have therefore been excluded, leaving the figures for the various categories as follows:

Mild cases:	vitamin C group	= 37 - 11 (excluded) = 26.,
	controls	= 37 - 9 (") = 28.
Moderate " :	vitamin C group	= 13 - 6 (") = 7 ,
	controls	= 13 - 0 (") = 13.
Severe cases:	vitamin C group	= 8,
	controls	= 8.

The classification of these cases into mild, moderate and severe was made by the medical superintendent; and the discharging of the patients was arranged by a colleague who did not know whether any particular patient had received ascorbic acid.

Details/

Details of the mild cases are given below in tables XII and XIII. The moderate and severe cases will be considered later.

Table XII.

Mild cases of diphtheria treated with vitamin C.

No.	Sex.	Age in yrs.	Description.	Dosage of serum in thousands of units.	Duration of illness in days.	Complications, and sequelae.
1	M	13	(F & L)1	30	42	Ciliary paresis.
2	F	25	F 1	6	29	-
3	M	2	F 1	10	51	Late adenitis.
4	M	4	F 1	6	43	-
5	F	8	F 1	4	29	-
6	F	10	F 1	4	30	-
7	F	20	F 1	8	36	-
8	F	5	F 1	8	40	-
9	F	2	F 1	8	31	-
10	F	11	F 1	8	49	-
11	F	21	F 1	4	28	-
12	F	11	NP 1	12	38	-
13	F	3	F 1	4	32	-
14	F	21	F 1	8	36	-
15	F	11	F 1	8	48	-
16	M	5	F 1	8	39	-
17	F	25	F 1	4	51*	-
18	M	6	F 1	10	37	-
19	M	5	F 1	10	42	-
20	F	19	F 1	6	51°	-
21	M	8	F 1	6	32	-
22	F	15	L 1	10	43	-
23	M	6	F 1	8	36	-
24	M	6	F 1	8	44	-
25	M	12	F 1	5	37	-
26	F	2	F 1	30	29	-

Table XIII.

Mild cases of diphtheria used as controls.

No.	Sex.	Age in yrs.	Description.	Dosage of serum in thousands of units.	Duration of illness in days.	Complications.
1	F	14	(F & L)1	10	42	-
2	F	19	F 1	6	33	-
3	F	26	F 1	4	37	-
4	M	5	F 1	8	44	-
5	F	17	(F & L)1	4	38	-
6	F	17	F 1	8	40	-
7	M	5	F 1	4	33	-
8	M	3	F 1	20	44	-
9	F	5	F 1	8	37	-

(Continued on next page.)

* Persistent +ve swabs. ° Intercurrent pyelitis.

Table XIII (continued).

No.	Sex.	Age.	Description.	Dosage.	Duration.	Complications.
10	F	8	F 1	5	26	-
11	F	26	AN 1	4	19	-
12	F	39	F 1	6	34	-
13	F	20	F 1	8	34	-
14	F	2	F 1	6	35	-
15	M	7	F 1	5	31	-
16	M	3	AN 1	4	28	-
17	M	20	F 1	10	54	-
18	F	7	F 1	6	37	-
19	F	8	F 1	4	26	-
20	M	5	F 1	8	44	-
21	M	7	F 1	5	32	-
22	M	4	F 1	10	37	-
23	F	16	F 1	10	37	-
24	F	23	F 1	4	26	-
25	F	19	F 1	8	40	-
26	M	13	F 1	8	44	-
27	M	6	F 1	8	37	-
28	M	3	F 1	20	44	-

Comparison of the mild cases.

The two series of mild cases are comparable in respect of age, as the following classification will show:

Age.	Vitamin C cases.	Controls.
Over 20 years.	4	4
10 - 20 "	9	9
6 - 9 "	5	6
4 & 5 "	4	5
2 & 3 "	4	4
	26	28.

Although all the cases were mild, there is some dis--parity between the groups in respect of degree of mildness. Classification according to the dose of antitoxin prescribed by the medical superintendent of the hospital reveals that the controls had a larger number of very mild cases:

Dose of serum.	Vitamin C cases.	Controls.
10,000 units or more.	7	6
8,000 "	9	8
6,000 " or less.	10	14
	26	28.

To obviate any possibility of an error arising from the preponderance of very mild cases among the controls, the comparison between the patients treated with ascorbic acid and the patients used as controls will be made in three separate groups, - cases that received at least 10,000 units of antitoxin, cases that received 8,000 units, and cases that received not more than 6,000 units.

Total/

	Total cases.	Complications, &c.	Average stay in hospital.	Median stay in hospital.
Vitamin C cases that received at least 10,000 units.	7	2	40.3 days.	42 days.
Similar controls.	6	0	43. "	43 " .
Vitamin C cases that received 8,000 units	9	0	39.9 "	39 "
Similar controls.	8	0	40. "	40 " .
Vitamin C cases that received 6,000 units or less.	10	0	36.2 "	32 "
Similar controls.	14	0	31.1 "	32.5 " .

There were no fatalities among these 54 cases, and complications were far too infrequent to allow of the drawing of even tentative conclusions. As for duration of illness, it will be observed that, in each comparable section, the median duration of stay in hospital is half to one day longer in the controls than in the treated cases, while the average is longer in the controls of two sections but shorter in the controls of the third section. In this third section the average for the vitamin C cases is con-siderably distorted by two patients (No. 17 and No. 20 in Table XII), so that the median is probably a truer guide.

For mild diphtheria, then, the results are very similar to those found in tonsillitis: the duration of illness in the cases treated with ascorbic acid appears to be a shade less than the duration in comparable controls, but the difference is so slight that it would probably be necessary to study several hundreds of cases before one could decide whether the difference was significant or due to chance.

Moderate and severe cases.

Details of the 20 moderate cases and of the 16 severe cases are given in Tables XIV and XV.

Table/

Table XIV.

Moderate and severe cases of diphtheria treated with vitamin C.

No.	Sex.	Age in yrs.	Description.	Dosage of serum in thousands of units.	Duration of illness in days.	Complications and sequelae.
1	M	2	NP 2	30	50	Tachycardia.
2	M	11	F 2	20	48	-
3	M	5	NP 2	20	108	Palatal, pharyngeal and diaphragmatic pareses.
4	M	1	F 2	20	49	-
5	M	3	(NP&L)2	20	56	-
6	F	3	F 2	20	55	-
7	M	4	F 2	20	38	-
A	F	16	NP 3	50	68	-
B	F	7	NP 3	50	76	-
C	M	3	NP 3	130	Died	on 8th. day in hospital.
D	F	9	NP x*	100	94	Heart failure; palatal, ciliary & facial pareses.
E	F	5	NP 3	100	72	-
F	M	6	NP 3	50	70	-
G	F	16	NP 3	100	68	-
H	M	16	NP 3	100	70	- .

* A haemorrhagic case which made a complete recovery.
" x " signifies, " hopeless prognosis ".

Table XV.

Moderate and severe cases of diphtheria used as controls.

No.	Sex.	Age in yrs.	Description.	Dosage of serum in thousands of units.	Duration of illness days.	Complications and sequelae.
1	M	3	(F & L)2	30	56	-
2	M	5	F 2	16	48	-
3	F	3	NP 2	16	47	-
4	F	11	F 2	20	47	-
5	F	27	(F & L)2	20	58	-
6	M	3	F 2	20	44	-
7	M	15	NP 2	20	Died	on 9th. day in hospital.
8	M	5	F 2	16	38	Strabismus.
9	M	3	NP 2	30	54	-
10	F	14	F 2	16	45	-
11	F	7	F 2	28	66	-
12	M	26	NP 2	30	42	-
13	M	41	NP 2	20	46	-
A	F	6	NP 3	100	Died	on 7th. day in hospital.
B	F	2	NP 3	100	Died	on 3rd. day " "
C	F	6	NP 3	50	Died	on 3rd. day " "
D	M	9	NP 3	46	67	Ciliary paresis.
E	M	4	NP 3	100	82	Late adenitis.
F	M	59	(NP&L)3	50	+	
G	F	4	NP 3	50	Died	on 2nd. day in hospital.
H	F	11	NP 3	100	Died	on 9th. " " "

+ Died /

+ Died of intercurrent lobar pneumonia on 35th. day of diphtheria.

- -

Comparison of moderate and severe cases.

As compared with the moderate cases which received vitamin C, the moderate controls are older (five of them being older than the oldest child in the treated group), and contain a larger proportion of relatively mild cases: four of the thirteen controls required only 16,000 units of antitoxin. Hence comparison of the two moderate groups has a very limited value. The main points of difference, for what they are worth, are as follows:-

	Total cases.	Deaths.	Sequelae.	Average duration in days.	Median duration in days.
Moderate vitamin C cases	7	0	2	57.7	50
Moderate controls	13	1	1	49.3	47 .

The two groups of toxic cases are quite well balanced in respect of dosage of serum: five treated cases and four controls had 100,000 units, while the others received 50,000 units. They are also fairly comparable in respect of age, although the control series contains one elderly adult.

The main points concerning the severe cases can be summarised thus:-

	Total cases.	Deaths.	Complications and sequelae.	Average duration in survivers.	Median duration in survivers.
Severe vitamin C cases	8	1	1	74. days.	70 days
Severe controls	8	5°	2	74.5 "	74.5 " .

One cannot, of course, form definite conclusions from sixteen severe cases; but the mortality rates (12.5 % and 62.5 % respectively) and the incidence of complications in survivers (14.29 % and 66.67 % respectively) show such marked differences as to make a further investigation eminently desirable.

Experiment /

- -

° The man who died of intercurrent lobar pneumonia is not included among the deaths.

Experiment II - Parenteral administration of vitamin C.

As a result of Experiment I the writer felt that mild cases did not merit further investigation, but that severe cases deserved further study (in view of the apparently beneficial results of ascorbic acid therapy in the small number of severe cases previously described), and that the second experiment might profitably include moderate cases also (in view of the fact that the moderate groups of Experiment I were not really comparable).

It seemed likely that, if orally administered vitamin C - which might or might not be absorbed from the alimentary tract - was of value in severe diphtheria, then, a fortiore, intravenous injection of the vitamin would be beneficial; and conversely, if intravenous ascorbic acid proved to be useless, oral treatment with the vitamin would probably be useless. Accordingly, the writer decided to give vitamin C parenterally, and - in view of the large doses tried by some of the German workers (see Part 1) - to use a dosage of such magnitude that any negative results could not possibly be attributed to inadequacy of amounts given. The quantity selected was, for a patient over the age of eight years, 500 mg. daily for ten days, and for a younger child 250 mg. daily for the same period.

Selection of cases.

To eliminate errors due to the presence of another variable, namely, dosage of antitoxin, it was decided to give vitamin C to every second case that received 100,000 units of serum, to every second case that received 50,000 units, and to every second case that received 30,000 units.

It was originally intended that the 5,000 mg. of vitamin C (or 2,500 mg. for young children) should be given in ten daily intravenous injections. However, as some of the patients admitted at the beginning of the experiment had very " difficult " veins, the plan was altered: each patient was given intravenous injections of 500 mg. or 250 mg. (according to age) for three days, and intramuscular injections of the same quantities of the vitamin for the next seven days.

In all, eighteen severe cases of diphtheria and six moderate cases were investigated. Details of the individual patients are recorded in Tables XVI and XVII on next page.

Table /

Table XVI.

Cases of diphtheria given ascorbic acid parenterally.

No.	Sex.	Age in yrs.	Dosage of serum in thousands of units.	Complications or Sequelae.	Duration of illness in days.
1	M	9	100	Palatal paresis.	69
2	F	4½	100	Died on 10th. day of disease (3rd. day in hosp.)	
3	F	11	100	-	108
4	M	14	100	-	69
5	M	4½	50	Strabismus and Palatal Paresis.	70
6	M	5	50	-	54
7	F	2 11/12	50	-	74
8	M	21	50	Palatal paresis	63
9	M	10	50	-	85
10	F	9	30	-	56
11	M	2	30	Palatal paresis	86
12	M	7	30	Palatal paresis	74 .

Table XVII.

Cases of diphtheria used as controls.

No.	Sex.	Age in yrs.	Dosage of serum in thousands of units.	Complications or Sequelae.	Duration of illness in days.
1	M	6	100	Palatal paresis, Died on 12th. day of disease (11th. day in hosp.)	
2	F	10	100	-	70
3	M	6	100	Died on 7th. day of disease (4th. day in hosp.)	
4	M	9	100	-	95
5	M	14	50	-	70
6	F	5	70*	Died on 12th. day of disease (8th. day in hosp.)	
7	F	1 4/12	50	-	73
8	M	4	50	Palatal and Ciliary pareses, Died on 110th. day of disease.	
9	M	8	50	-	103
10	M	6	30	-	60
11	F	3	30	Strabismus	73
12	F	2	30	Strabismus	67 .

* The 70,000 units in Case 6 includes 20,000 prior to admission.

It should perhaps be mentioned that the only fatality among the vitamin C patients - Table XVI, Case No. 2 - was a haemorrhagic case.

The /

The main points concerning the two groups may be tabulated thus:-

	Total cases.	Number who died.	No. who developed complic--ations.	Average duration of illness in survivers.	Median duration of illness in survivers.
a) Patients given 100,000 units of antitoxin.					
Vitamin C cases	4	1	1	82. days.	69 days.
Controls	4	2	1	82.5 "	82.5 "
b) Patients given 50,000 units.					
Vitamin C cases	5	0	2	69.2 "	70 "
Controls	5	2	1	82. "	73 "
c) All severe cases.					
Vitamin C cases	9	1	3	74. "	69.5 "
Controls	9	4	2	82.2 "	73 "
d) Moderate cases (30,000 units).					
Vitamin C cases	3	0	2	72. "	74 "
Controls	3	0	2	66.7 "	67 "
e) Total cases.					
Vitamin C cases	12	1	5	73.5 "	70 "
Controls	12	4	4	76.4 "	71.5 "

The results run roughly parallel with those of the first experiment: i.e. in the moderate cases (those that received 30,000 units of serum) there is little difference between the treated patients and the controls, but the toxic cases which were given ascorbic acid show a considerable lowering of the mortality rate as compared with the controls, - 1/9 as against 4/9.

Consideration of severe cases from both experiments.

If we now take all the toxic cases from Experiments I and II, and divide them into those who received vitamin C (irrespective of whether the vitamin was given orally or by injection) and those who were used as controls, we can form two groups, each containing 17 patients. These groups are comparable in respect of sex, - for there are nine males and eight females in each group; in respect of age, - for each contains six children of five years or under, ten older children/

children, and one adult; and in respect of dosage of serum, - for the vitamin C group comprises nine patients who were given 100,000 units and eight who received 50,000 units, while the control series contains eight who were given 100,000 units and nine who received 50,000. Moreover, since these two series represent a summation of small groups to which the alternate case method was invariably applied, differences due to changes in the virulence of the organism should be minimised.

In these two well balanced groups there is a very marked difference in the mortality rates: the death rate among the cases that received vitamin C is 2/17, or 11.76%, whereas among the controls it is 9/17, or 52.94%. The actual difference in probability of death in the two groups is 41.18%, or 2.86 times the standard error of the differ--ence. In other words, the difference is definitely significant.

Comparison of oral and parenteral administration.

It is never very satisfactory to compare groups of patients treated at different times; but, since the death rates and complication rates in the toxic control groups of the two experiments were not dissimilar (deaths = 5/8 and 4/9 respectively, and patients with complications = 2/8 and 2/9 respectively), it was felt that a comparison between the severe cases that received vitamin C orally and those that received it by injection might be attempted.

The eight toxic cases that were given 800 mg. of the vitamin by mouth included one that died and one that developed various complications, the latter case being a haemorrhagic diphtheria which made a complete recovery. The nine toxic cases that received 5,000 mg. parenterally included one that died (a haemorrhagic case) and three that developed complications.

It seemed probable, then, that the very high dosage employed in Experiment II was unnecessary, and that paren--teral administration (which is more laborious and much more expensive) possessed no marked advantage over treat--ment by the oral route. Nevertheless, since diphtheria is not infrequently characterised by initial nausea and sickness, it was felt that at least the first dose of the vitamin might well be given parenterally.

Accordingly, it was decided that, in the next exper--iment, patients would be given 500 mg. intravenously, and thereafter would receive their ascorbic acid by mouth.

Experiment /

Experiment III - Confirmatory.

Although the two experiments conducted at Edinburgh City Hospital had revealed a significant difference between the mortality rates of severe cases of diphtheria treated with vitamin C and those of severe cases that did not have the benefit of such treatment, a further investigation was deemed advisable, not so much to corroborate the general finding that ascorbic acid was of value, as to confirm the decision that the major portion of the vitamin could be given orally. Accordingly, at Kendray Isolation Hospital, Barnsley, during the winter of 1941-42, every alternate severe case and every alternate moderate case of diphtheria was treated with vitamin C, the dosage employed being 500 mg. intravenously (irrespective of patient's age) on admission, and 200 mg. or 100 mg. daily (according to whether the patient was over or under the age of eight years) for three days. The control patients received no supplement of vitamin C during the first week of their stay in hospital. In the second and later weeks all patients were given vitamin C orally in small quantities, sufficient to raise the ascorbic acid level of war-time diets to the more generous standard of normal times.

Possible fallacies.

In a small hospital, where cases admitted during the morning or afternoon were seen by the writer but where evening duty was taken in rotation by four members of the staff of the public health department, one could not hope to achieve the same standard of uniformity of dosage as is obtained in larger institutions; but, while an isolated case might receive an amount of serum slightly disproport--ionate to its severity, there is no reason to suppose that such occasional slight disproportions would upset the balance between the groups of cases.

A fact more likely to lead to error is that the discharging of the patients was in the hands of the writer, who might be unconsciously biased by his knowledge of whether any particular case had been treated with vitamin C. To obviate any fallacy from this cause, "Duration of illness" has been omitted from the criteria used in compar--ing the groups: treated cases and controls have been compared only in respect of incidence of deaths and of frequency of complications and sequelae.

Details of cases.

The alternate case method was applied to moderate cases which received about 40,000 units of antitoxin, and to severe cases that were given about 60,000 units. These categories correspond with Edinburgh cases receiving 30,000 units and 50,000 to 100,000 units respectively, the dosage/

dosage used being generally a shade higher than that employed in Edinburgh, because south Yorkshire seems to harbour a particularly virulent strain of C. Diphtheriae.

Details of individual cases are recorded in Tables XVIII and XIX.

Table XVIII.

Cases of diphtheria treated with vitamin C.

No.	Sex.	Age in yrs.	Description*	Dosage of serum in thousands of units.	Complications, &c.
1	F	11	F 3	60	-
2	F	12	F 3	60	-
3	F	4	NP 3	80	Otitis, unilateral.
4	F	4	F 3	60	-
5	F	2	F 3	60	-
6	F	6	F 3	60	Cardiac failure.
7	F	1 10/12	F 3	80	-
8	F	11	F 3	60	-
9	M	5	F 3	60	Neuritis.
10	F	10	NP 3	80	-
11	F	10	NP 3	60	-
12	M	8	F 3	60	-
A	F	4	L 2	40	-
B	M	9	F 2	40	-
C	M	14	NP 2	40	-
D	F	16	F 2	40	-
E	F	4	F 2	50	-
F	F	2	F 2	40	-

Table XIX.

Cases of diphtheria used as controls.

No.	Sex.	Age in yrs.	Description*	Serum (in thousands of units.)	Complications, &c.
1	M	13	NP 3	60	Bradycardia.
2	F	11	NP 3	60	Palatal paresis; <u>died</u> on 43rd. day.
3	F	12	F 3	80	Cardiac failure.
4	F	11	F 3	90	Palatal and ciliary paresis.
5	F	12	F 3	80	-
6	F	8	NP 3	80	<u>Died</u> on 1st. day in hosp.
7	M	24	F 3	60	-
8	F	20	F 3	60	-
9	F	18	F 3	60	-
10	M	7	F 3	60	-
11	M	5	F 3	60	-
12	M	2	F 3	60	Cardiac failure.
A	F	7	F 2	40	Facial & palatal paresis.
B	F	7	F 2	40	-
C	M	6	F 2	40	-
D	F	9	F 2	50	Cardiac failure.
E	F	5	F 2	40	Cardiac failure & strabismus.
F	F	14	F 2	40	Palatal paresis.

* The /

The contractions used in the description of cases - "F 2", "F 3", etc. - have the same significance as in the earlier experiments.

Comparison of the groups.

There are more young children among the vitamin C cases than among the controls, - a fact which should certainly not incline the balance in favour of the treated cases. In other respects the series are quite comparable.

Some of the main points may be tabulated thus:-

	Total cases.	Number who died.	No. with sequelae.	No. who had an uncomplicated convalescence.
a) Severe				
Vitamin C cases	12	0	3	9
Controls	12	2	5°	6
b) Moderate				
Vitamin C cases	6	0	0	6
Controls	6	0	4	2
c) Total				
Vitamin C cases	18	0	3	15
Controls	18	2	9°	8 .

° Including one patient who died later (and who is therefore counted in two separate columns).

In these two groups of 18 patients, the difference in the probability of recovery without sequelae is 38.89%, or 2.66 times the standard error; and the difference in the probability of death is 1.5 times the standard error.

Discussion of findings in all three experiments.

The 150 cases of diphtheria that were investigated comprised 54 mild, 38 moderate, and 58 severe cases. From these not inconsiderable numbers, it should be possible to determine whether ascorbic acid therapy serves any useful purpose.

A drug may be deemed of value in diphtheria if it shortens the period of illness, or if it reduces the frequency of sequelae and complications, or if it lowers the death rate. Let us apply these three criteria to treatment with vitamin C.

(1) Does administration of vitamin C shorten the period of illness ?

In Experiment I it was found that, for three separate/

separate groups of mild cases, the median duration of illness in cases treated with vitamin C was half to one day less than the median duration in the controls; and in the group of severe cases the median was 4½ days less in the treated cases than in the controls. The severe cases of Experiment II gave similar results: the median length of illness was 3½ days shorter in treated patients than in controls. There are therefore five groups of patients in which the median duration was slightly less in the vitamin C cases than in the controls; and in four of these five the average duration was also shorter in the treated cases, while in the fifth - the mild patients who received not more than 6,000 units of antitoxin - the average was distorted by two abnormal cases.

On the other hand, there are two groups - the mod--erate cases of Experiment I and of Experiment II - where the average duration and the median duration of illness were both longer in the treated cases than in the controls; but the moderate cases of Experiment I have been shown to be hardly comparable, and the moderate cases of Experiment II form an exceedingly small group.

On balance, the patients treated with ascorbic acid were discharged from hospital a little earlier than the ones who were not so treated, but the differences are so slight that only by a most extensive and prolonged invest--igation would it be possible to determine whether these differences were really significant.

The question, " Does administration of vitamin C shorten the period of illness ? ", cannot be answered with a definite affirmative.

(2) Does treatment with the vitamin reduce the incid--ence of complications or of sequelae ?

In the 150 cases studied, complications - as distinct from sequelae - were very infrequent: there were two compl--ications (one case of otitis and one of late adenitis) among the treated cases, and one (late adenitis) among the controls.

The percentage of patients who developed sequelae was 15.49 in the vitamin C cases, and 18.99 in the controls; and the total number of sequelae per hundred patients was 23.94 in the treated cases and 24.05 in the controls. A detailed consideration of the individual sequelae suggests, however, that there may be a difference between the vitamin C cases and the controls: while post-diphtheritic paralysis was equally common in treated and in untreated cases - 14 instances of paralysis occurred in each category -, there was a certain amount of disparity in the incidence of the most dangerous of the common sequelae, cardiac failure.

Out /

Out of 79 controls, five developed non-lethal heart failure (to say nothing, for the moment, of the twelve who died), whereas in the 71 cases treated with vitamin C there were - apart from the two patients who died - only three cases of cardiac failure.

It can be stated definitely that ascorbic acid therapy does not reduce the frequency of post-diphtheritic paresis. Whether it does anything to prevent cardiac (or circulatory) failure will - since this failure is the common cause of death in diphtheria and was responsible for all the deaths in the observed cases - be shown by the answer to question three.

(3) Does administration of ascorbic acid lower the death rate ?

A comparison of all the moderate and severe cases that received ascorbic acid with all the moderate and severe controls would be unfair to the treated patients for two reasons, - first, because the moderate controls of Experiment I have already been shown to be rather milder than the corresponding treated cases; and, second, because the disparity in numbers between the moderate controls of the first Experiment and their treated counterparts would cause the total group of controls to contain a higher percentage of moderate cases than the total group of treated cases. Nevertheless, let us make the comparison.

The moderate and severe cases treated with vitamin C consist of 29 toxic cases and 16 moderate cases, - i.e. 45 in all. The corresponding controls comprise 29 toxic cases and 22 moderate cases, - i.e. 51 in all. The mortality rate in the treated cases is 2/45, or 4.44 %, while in the controls it is 12/51, or 23.53 %. The standard error of the difference is $\pm$6.63. Hence the actual difference of 19.09 % is 2.85 times the standard error: so, even in this weighted comparison, the difference is significant.

If we disregard the moderate cases (which included no fatalities among treated cases and only one death among the controls), and contrast the 29 toxic cases that received vitamin C with the 29 toxic cases that did not have any such treatment, we find that the proportion of fatalities is 2/29 in the one group and 11/29 in the other, the actual difference being 3.05 times the standard error.

Even more convincing than these statistical calculations is a simple summary of the four groups in which deaths occurred:-

Description.	Vitamin C cases. Deaths/Cases.	Controls. Deaths/Cases.
Moderate cases, Exp. I	0/7	1/13
Severe " " I	1/8	5/8
Severe " " II	1/9	4/9
Severe " " III	0/12	2/12
Totals /		

	Vitamin C cases. Deaths/Cases.	Controls. Deaths/Cases.
Totals	2/36	12/42.

Administration of vitamin C, then, definitely lowers the mortality rate.

Dosage and method of administration.

The purpose of the investigations previously described was to determine whether treatment with vitamin C served any useful purpose. However, since the treated cases of the first experiment received 800 mg. orally, those of the second experiment 5,000 mg. parenterally, and those of the last experiment 500 mg. intravenously followed by 600 mg. orally, it may be possible to form a rough idea of the best route of administration and of the minimum adequate dose of the vitamin.

Comparison of groups of patients treated at different times and in different places is, of course, never really satisfactory. However, a consideration of the various control groups suggests that the virulence of the organism and the resistance of the host did not vary unduly:

Group.	Total cases.	Deaths.	Heart failure with recovery.
Toxic controls of Exp. I	8	5	0
" " " " II	9	4	0
" " " " III	12	2	3
Moderate " " " I	13	1	0
" " " " II	3	0	0
" " " " III	6	0	2.

The corresponding figures for the treated cases are:

Group.	Total cases.	Deaths.	Heart failure with recovery.
Toxic cases of Exp. I	8	1	1
" " " " II	9	1	0
" " " " III	12	0	1
Moderate " " " I	7	0	1
" " " " II	3	0	0
" " " " III	6	0	0 .

Despite the many possible fallacies inherent in a comparison of such groups, it may be claimed that the exceedingly high dosage of Experiment II is unnecessary: the patients of Experiment III made quite as good progress. On the other hand, the cases of Experiment I seem to be a shade less favourably placed. This may well be due to chance, or to the earliest cases investigated having happened to be a little more toxic; but, until further evidence is available, it is safer to assume that cases of Experiment I were given rather less than the optimum dosage./

dosage.

It appears, then, that a case of diphtheria will receive sufficient ascorbic acid if given 500 mg. intra--venously, followed by 300 to 600 mg. orally, according to age.

Ascorbic acid therapy in diphtheria, - Conclusions.

It is doubtful whether administration of vitamin C to mild cases of diphtheria serves any useful purpose other than that of remedying the state of hypovitaminosis C, - which state could be just as effectively remedied by the provision during convalescence of a diet containing abundance of fresh fruit and green vegetables.

In all severe cases of diphtheria, on the other hand, treatment with adequate dosage of vitamin C is eminently desirable: such treatment significantly reduces the mortality rate.

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SUMMARY AND CONCLUSIONS.

To ascertain whether the administration of ascorbic acid in certain infectious diseases was attended by any beneficial results, some 456 patients were investigated in Edinburgh City Hospital, and 52 patients in Kendray Isolation Hospital, Barnsley, were studied in subsequent corroborative experiments. Immediately after admission to hospital, cases of each disease were allocated - on the alternate case principle - to one or other of two groups. These groups received similar medicaments and diet, except that Group I was given synthetic vitamin C. The main findings are mentioned in the following summary.

1. Influenza.

108 adult males, admitted during an epidemic of a fairly mild type of influenza, were divided into two groups, comparison of which revealed no serious discrep--ancy in respect of initial severity, age distribution, etc. Members of Group I received 400 mg. of vitamin C during their first two days in hospital, while members of Group II were used as controls.

Although the patients in Group I had fewer relapses and /

and complications, and a shorter average length of illness, the observed differences were not of such magnitude as to be statistically significant: the difference in probability of recovery without complications was 2.16 times the standard error of the difference, and the difference in average dur--ation of illness was only 1.66 times the standard error.

The dosage of vitamin C used in this experiment was selected in the light of Falke's estimates of the amount of the vitamin metabolised in pyrexia, but investigation of the urinary excretion of patients in Group I suggested that the amount of ascorbic acid administered had been insufficient to remedy the state of hypovitaminosis C. However, since influenza is seldom treated in isolation hospitals, no opportunity was found of repeating the experiment with a higher dosage of the vitamin.

2. Cerebrospinal Meningitis.

(a) Oral administration of the vitamin.

For 120 consecutive admissions each alternate case was offered 600 mg. of ascorbic acid by mouth during the first four days in hospital. As some patients were too sick to retain the vitamin, a standard of exclusion equally applicable to both groups had to be devised, and for this purpose ability to tolerate sulphapyridine given by mouth was selected (because retention of the latter drug obviously implies ability to retain ascorbic acid). In all, 42 members of Group I and 46 controls from Group II were able to take drugs orally.

There was no real disparity between the groups in respect of duration of illness, but at first glance it looked as though the vitamin C cases had a materially lower death rate and a smaller incidence of complications. Scrutiny of the two groups revealed, however, that the controls included a disproportionate number of infants and elderly persons. Death rates and complication rates were therefore standardised, and it was found that, although the differences remained in favour of the group treated with vitamin C, these differences were far below the level of significance: the difference between the standardised death rates was 1.05 times the standard error, whilst that between the standardised complication rates was 1.22 times the standard error.

(b) Parenteral administration of the vitamin.

After the oral experiment had been in progress for a short time, it was decided that cases excluded from Group I (on the ground of inability to tolerate sulphapyridine orally) should receive 500 mg. of vitamin C intramuscularly, while similar exclusions from Group II should be used as controls /

controls. The injections were given approximately 24 hours after each patient had been admitted to hospital, and they were, of course, additional to any portion of the first day's oral dosage that the patient had managed to retain. Twelve cases received intramuscular injections of ascorbic acid, and thirteen cases fell into the control series.

Contrary to the investigator's expectation, the differences between the two groups were dramatic: in the vitamin C cases the death rate was very little more than half of the death rate among the controls, while the percentage of recoveries without complications was almost twice that of the control group.

In view of these remarkable differences, a further trial of parenteral ascorbic acid was judged to be most desirable. Accordingly, 30 consecutive cases admitted to hospital suffering from meningitis were divided into the usual two groups, and the patients in Group I received 1500 mg. of vitamin C intramuscularly during their first three days in hospital (500 mg. daily), whilst members of Group II served as controls. Owing to treatment being commenced before laboratory reports were to hand, two of the Group I cases had to be excluded as pneumococcal, not meningococcal. However, the results in the remaining cases were sufficiently striking: the 13 cases treated with vitamin C included no fatalities and no complications, as compared with three deaths and three complications in the control group. Despite the smallness of the numbers, the difference in the probability of an uncomplicated convalescence was found to be significant.

The 25 cases that received vitamin C parenterally have been compared as a group with the 28 controls; to eliminate any possible error due to differences in the age distribution, a comparison in age-groups has been made; and (since a study of the controls used at various times showed that, during the portion of the epidemic in which these experiments were conducted, the disease did not undergo any marked change in its severity) the cases that received vitamin C by injection have been compared with all the other cases of the disease, - i.e. oral vitamin C cases, controls of oral cases, and controls for parenteral cases. Every single comparison emphasised the favourable position of the cases treated with injections of ascorbic acid.

To " make assurance double sure " a confirmatory experiment was carried out at Kendray Hospital, each alternate admission receiving 1500 mg. of the vitamin in three daily injections. The eight cases in Group I included no fatalities and only two with transient complications, as contrasted with one death and four complications - three /

- three of them lasting - among the eight controls. So these cases, although not numerous enough to be significant when considered by themselves, corroborate the remarkable results of the Edinburgh experiments.

3. Streptococcal Tonsillitis, with or without a rash.

For a period of eight days, 16 cases of tonsillitis due to Str. Pyogenes were given 50 or 100 mg. of ascorbic acid each day, orally, while 16 similar cases were used as controls. Such differences as were found were in favour of the vitamin C cases but were not statistically signif--icant: the average duration of inflammation of the tonsils was 1.44 days more in the controls than in the treated cases, the standard error of the difference being ± 1.34; and the average period of hospitalisation was 1.25 days longer in the controls, the standard error of the difference being ± 1.54.

Of 40 consecutive cases of scarlet fever every alternate case was given ascorbic acid, the dosage used being the same as in simple tonsillitis. There was, in favour of the treated cases, a slight disparity in the average duration of tonsillar inflammation, but it amounted to only 1.12 times the standard error of the difference; and no other points of difference between the groups were noted.

4. Post-Influenzal Pneumonia.

Every alternate case admitted with bronchopneumonia following influenza received 200 mg. of ascorbic acid by mouth each day for three days. Lack of patients brought the experiment to a close before an adequate number of cases had been investigated. However, consideration of the 14 cases studied suggests that vitamin C therapy does not produce any marked effects.

5. Diphtheria.

(a) Oral administration of vitamin C.

Cases of diphtheria were classified on admission as mild, moderate, and severe cases. In each of these three categories every alternate patient was given ascorbic acid for eight days, the daily dose being 50 or 100 mg., accord--ing to age.

The 54 mild cases investigated included no fatalities and only two with sequelae. The duration of illness was, on the average, a shade less in the treated cases than in the controls, but the difference was very far below the level of significance. The two groups of moderate cases - 20 in all - could not legitimately be compared owing to disparities in age-distribution and in dosage of antitoxin. The /

The two groups of severe cases were well balanced in respect of dosage of serum, age of patients, etc.; yet the 8 cases that received vitamin C included only one fatality, whereas the 8 controls included five who died.

It was therefore felt that, while mild cases needed no further study, moderate and severe cases required more inves--tigation.

(b) Parenteral administration of the vitamin.

It was decided that, to ensure absorption, the vitamin should be given intravenously or intramuscularly, in large dosage. To eliminate any errors due to the presence of a second variable - namely, amount of antitoxin - the alternate case principle was applied to all patients who received 30,000 units of serum, to all who received 50,000 units, and to all who received 100,000 units. Every second case in each of these categories was given 2,500 mg. or 5,000 mg. of ascorbic acid, according to age, during the first ten days in hospital.

The twenty-four patients investigated were compared class by class. In the 30,000 class there was little dis--crepancy between the treated cases and the controls. In the other two classes, however, there were marked differences in the mortality rates; and when the 50,000 units and 100,000 units groups were considered together it was found that the difference in probability of death in the severe cases that received vitamin C and in the severe controls was 2.66 times the standard error of the difference, - i.e. the difference was significant.

After discussion of the relative merits of the small oral dose used in Experiment (a) and the very large parenteral dose employed in Experiment (b), a compromise dosage of 500 mg. intravenously, followed by 300 to 600 mg. orally, was suggested.

(c) Confirmatory experiment.

In a confirmatory experiment the compromise dosage mentioned above was given to every alternate severe case and to every alternate moderate case. The twelve severe cases that received ascorbic acid included none that died and only three that developed complications or sequelae, whereas the twelve severe controls included two who died and four who suffered from sequelae. In the moderate groups there were no deaths, but, whilst the six treated cases recovered without any after effects, four of the six controls developed sequelae.

In a discussion of the entire 150 patients who were investigated it has been shown that administration of vitamin C significantly lowers the mortality rate, and it has been suggested that it acts mainly by reducing the frequency, and the /

the severity, of diphtheritic heart failure. On the other hand, treatment with vitamin C does not decrease the incid--ence of post-diphtheritic paralysis.

Conclusions.

1. In cerebrospinal meningitis the value of intramuscular injections of ascorbic acid is proved beyond shadow of doubt. While it is not for a moment suggested that treatment with vitamin C should replace sulphapyridine or sulphathiazole therapy, the vitamin is a most useful adjuvant: injection of 1500 mg. during the first three days of illness causes a dramatic drop both in the percentage of deaths and in the incidence of complications and sequelae.

2. Owing to gastro-intestinal upset, oral administration of ascorbic acid is often impracticable in cases of cerebro--spinal meningitis; and, even in cases which can tolerate drugs given by mouth, it is doubtful whether any real benefit is conferred by oral administration of moderate doses of the vitamin.

3. In severe faucial or nasopharyngeal diphtheria treatment with adequate amounts of the vitamin significantly reduces the mortality rate. An adequate dose for a severe case of diphtheria is, - 500 mg. intravenously, with (or immediately after) the antitoxin, followed by 600 mg. orally during the next three days.

4. Since the dosage just mentioned gives satisfactory results in toxic diphtheria, the use of the parenteral route after the first day of treatment appears to be unnecessary and - in view of the relative costs of tablets and sterile ampoules - undesirable.

5. In mild diphtheria, streptococcal tonsillitis, and scarlet fever oral administration of the vitamin does not produce very marked results: such effects as are occasioned by ascorbic acid therapy are certainly in the direction of shortening the period of illness and the duration of faucial inflammation, but the benefits are so trivial that it is doubtful whether the giving of synthetic ascorbic acid in these diseases is a justifiable expense. It seems probable that, under the conditions of normal times, all that is necessary is the provision of sufficient fruit drinks during the acute stage and of ample fresh fruit and green vegetables during convalescence.

6. In cases of influenza a study of the urinary excretion of ascorbic acid proves that 400 mg. of the vitamin (or Falke's suggested dosage of 100 mg. for each day of pyrexia) is insufficient to remedy the state of hypovitaminosis C. Since /

Since treatment with even this inadequate dosage has been found, in a comparison of only fifty-four treated and fifty--four untreated cases, to cause a diminution in the incidence of complications and relapses sufficiently marked to approach the level of statistical significance, it is suggested that - until some subsequent investigator definitely proves or disproves the value of ascorbic acid in influenza - cases of that disease should be given about 200 mg. of the vitamin daily for the first three days of illness.

In brief, then, parenteral administration of vitamin C should form part of the treatment of cerebrospinal mening--itis, the drug should be given orally and intravenously in severe diphtheria, and it is probably desirable to give the vitamin orally to cases of influenza; but in mild diphtheria, scarlet fever and tonsillitis the provision of synthetic ascorbic acid appears to be an unjustifiable expense.

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